

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

10/066,007

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

L1 4 S ASTAXANTHIN (W)SYNTHASE?
L2 0 S ASTAXANTHIN (W)SYNTHETASE?
L3 6416 S ASTAXANTHIN
L4 24046 S "AST"
L5 194127 S SYNTHETASE?
L6 0 S L3 (W)L5
L7 39 S L3 AND L5
L8 24 DUP REM L7 (15 DUPLICATES REMOVED)
L9 41050 S REACTION (A) CENTER
L10 0 S L8 AND L9
L11 153 S L5 AND L9
L12 371127 S REDUCTASE?
L13 21 S L11 AND L12
L14 19 DUP REM L13 (2 DUPLICATES REMOVED)
L15 7593 S P450 REDUCTASE?
L16 0 S L9 AND L15
L17 0 S L4 AND L9
L18 59612 S "BETA-CAROTENE"
L19 64 S L4 AND L18
L20 0 S L9 AND L19
L21 0 S L1 AND L9
L22 3 DUP REM L1 (1 DUPLICATE REMOVED)
L23 93 DUP REM L11 (60 DUPLICATES REMOVED)
E HOSHINO T/AU
L24 4600 S E3
E KAZUYUKI O/AU
L25 6 S E3
E YUTAKA S/AU
L26 12 S E3
L27 4617 S L24 OR L25 OR L26
L28 19 S L5 AND L27
L29 13 DUP REM L28 (6 DUPLICATES REMOVED)

=>

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=> s astaxanthin (w)synthase?
L1 4 ASTAXANTHIN (W) SYNTHASE?

=> s astaxanthin (w)synthetase?
L2 0 ASTAXANTHIN (W) SYNTHETASE?

=> s astaxanthin
L3 6416 ASTAXANTHIN

=> s "AST"
L4 24046 "AST"

=> s synthetase?
L5 194127 SYNTHETASE?

=> s l3 (w)l5
L6 0 L3 (W) L5

=> s l3 and l5
L7 39 L3 AND L5

=> dup rem l7
PROCESSING COMPLETED FOR L7
L8 24 DUP REM L7 (15 DUPLICATES REMOVED)

=> d 1-24 ibib ab

L8 ANSWER 1 OF 24 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN
DUPLICATE 1

ACCESSION NUMBER: 2005-22605 BIOTECHDS

TITLE: New genetically engineered bacterium having a disruption in
one or more gdhA, gpmB aceE, ppc, talB, fdhF, yjiD, hnr or
yjfP genes, useful for overexpressing genes impacting
carotene biosynthesis and enhancing carotenoid production;
involving vector-mediated gene transfer and expression in
host cell

AUTHOR: STEPHANOPOULOS G; ALPER H S; JIN Y

PATENT ASSIGNEE: MASSACHUSETTS INST TECHNOLOGY

PATENT INFO: WO 2005062923 14 Jul 2005

APPLICATION INFO: WO 2004-US43295 23 Dec 2004

PRIORITY INFO: US 2004-609246 14 Sep 2004; US 2003-531996 24 Dec 2003

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2005-522448 [53]

AB DERWENT ABSTRACT:

NOVELTY - A genetically engineered bacterium comprising a disruption in

one or more *gdhA*, *gpmB*, *aceE*, *ppc*, *talB*, *fdhF*, *yjiD*, *hnr* or *yjfP* genes, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for the following: (1) a carotenoid-producing bacterium, comprising one or more inhibitors of *gdhA*, *gpmB*, *aceE*, *ppc*, *talB*, *fdhF*, *yjiD*, *hnr* or *yjfP* gene activity, or their combination, where use of the one or more inhibitors results in enhanced carotenoid production in the bacterium, as compared to wildtype; (2) a method for production of carotenoids, comprising genetically disrupting a *gdhA*, *gpmB*, *aceE*, *ppc*, *talB*, *fdhF*, *yjiD*, *hnr* or *yjfP* gene, or their combination in a cell comprising genes involved in the carotenoid biosynthetic pathway, and isolating carotenoids from the cell, and producing carotenoids; (3) a method for enhanced production of carotenoids, comprising contacting a cell comprising genes involved in the carotenoid biosynthetic pathway with one or more inhibitors of *gdhA*, *gpmB*, *aceE*, *ppc*, *talB*, *fdhF*, *yjiD*, *hnr* or *yjfP* gene expression or function, or their combination, and isolating carotenoids from the cell, and enhancing production of carotenoids; (4) a cell genetically engineered to over-expresses one or more *dxs*, *idi*, *yjiD*, *rpoS*, *torC*, *appY*, *ydgK*, *yeiA*, *yedR*, *tort*, *arcB*, *yggT*, *purDH*, or *yfjN* genes; (5) a method of determining optimized production of a metabolite, comprising constructing a flux balance analysis model, applying constraints to the flux balance analysis model comprising maximizing cell growth yield subject to a minimization of metabolic adjustment alteration, conducting in silico gene knockout simulations for all genes in the organism's genome, where a flux profile comprising a deletion of a knocked out gene from the stoichiometry matrix is calculated for the gene knockout simulations, and conducting in silico gene knockout simulations of all possible pairs or triplets of genes in the organism's genome, where a flux profile comprising the deletions of genes from the stoichiometric matrix is calculated for the gene knockout simulations, and selecting gene targets on the basis of enhanced flux profiles as a measure of optimal production of the metabolite, and determining optimized production of a metabolite; (6) a method of identifying genes involved in optimized production of a carotenoid, comprising constructing a flux balance analysis model, applying constraints to the flux balance analysis model comprising maximizing cell growth yield subject to a minimization of metabolic adjustment alteration, conducting in silico gene knockout simulations for all genes in the organism's genome, where a flux profile comprising a deletion of a knocked out gene from the stoichiometry matrix is calculated for the gene knockout simulations, conducting in silico gene knockout simulations of all possible pairs or triplets of genes in the organism's genome, where a flux profile comprising the deletions of genes from the stoichiometric matrix is calculated for the gene knockout simulations, selecting gene targets on the basis of enhanced flux profiles as a measure of optimal production of the metabolite, contacting cells comprising genes involved in the carotenoid biosynthetic pathway with a library of transposon mutagenized genes, selecting cells with enhanced carotenoid synthesis or production, and identifying mutagenized sequences, and inhibiting or abrogating gene expression of the gene targets identified, and their combinations, in a cell comprising genes involved in the carotenoid biosynthetic pathway, and determining carotenoid production in the cell, and identifying genes involved in optimized production of a carotenoid; and (7) a genetically engineered cell with enhanced lycopene production, where the cell is genetically disrupted for a gene or genes identified in the method of (6), and where the cell is suppressed or inhibited for expression of a gene or genes identified in the method of (6).

BIOTECHNOLOGY - Preferred Bacterium: The genetically engineered bacterium comprises a disruption in the genes *gdhA* and *gpmB*, genes *gdhA* and *aceE*, genes *gdhA* and *ppc*, genes *gdhA* and *fdhF*, or genes *gdhA* and *talB*, and further comprises a disruption in the *fdhF* or *talB* gene, and additionally comprises a disruption in the genes *gdhA*, *aceE* and *fdhF*, genes *gdhA* and *yjfP*, genes *gdhA*, *aceE* and *yjfP*, genes *gdhA*, *aceE*, *fdhF* and *yjfP*, genes *gpmB* and *yjiD*, genes *gdhA*, *aceE*, and *yjiD*, genes *gdhA*, *gpmB* and *yjiD*, genes *gdhA*, *aceE*, *hnr* and *yjfP*. The genetically engineered bacterium over-expresses *dxs*, *idi*, *yjiD*, *rpoS*, *torC*, *appY*, *ydgK*, *yeiA*, *yedR*, *tort*, *arcB*, *yggT*, *purDH*, *yfjN*, or their combination. The bacterium belongs to the *Escherichia*, *Methylobacter*, *Methylococcus*, and *Methylosinus* *Salmonella*, *Erwinia*, *Haematococcus*, *Rhodobacter*,

Myxococcus, Corynebacteria, Pseudomonas or Bacillus genus. The bacterium further comprises an isopentenyl pyrophosphate isomerase (Idi), a farnesyl pyrophosphate synthetase (IspA), a geranyltranstransferase, an octoprenyl pyrophosphate synthase (IspB), a geranylgeranyl pyrophosphate (GGPP) synthase (CcrtE), a phytoene synthase (CrtB), a phytoene desaturase (CrtI), a lycopene cyclase (CrtY), a beta-carotene hydroxylase (CrtZ), a zeaxanthin glucosyl transferase (CrtX), a beta-carotene ketolase (CrtO), or their combination. The one or more inhibitors in the carotenoid-producing bacterium comprise a nucleic acid. The presence of the one or more inhibitors in the bacterium results in enhanced carotenoid production of between 3 -50%, and suppresses or abrogates *gdhA* and *gpmB*, *gdhA* and *aceE*, *gdhA* and *ppc*, *gdhA* and *fdhF*, *gdhA* and *talB*, *fdhF*, *talB*, *gdhA*, *aceE* and *fdhF*, *gdhA* and *yjfp*, *gdhA*, *aceE* and *yjfp*, *gdhA*, *aceE*, *fdhF* and *yjfp*, *gpmB* and *yjiD*, *gdhA*, *aceE*, and *yjiD*, *gdhA*, *gpmB* and *yjiD*, *gdhA*, *aceE*, *hnr* and *yjfp*, *gdhA*, *aceE*, *hnr*, *yjfp* and *yjiD*, and/or *gdhA*, *gpmB* and *yjfp* gene expression or function. The bacterium also over-expresses *dxs*, *idi*, *yjiD*, *rpoS*, *torC*, *appY*, *ydgK*, *yeiA*, *yedR*, *tort*, *arcB*, *yggT*, *purDH*, *yfjN*, or their combination. The bacterium belongs to the *Escherichia*, *Methylobacter*, *Methylococcus*, and *Methylosinus Salmonella*, *Erwina*, *Haematococcus*, *Rhodobacter*, *Myxococcus*, *Corynebacteria*, *Pseudomonas* or *Bacillus* genus.

The bacterium further comprises an isopentenyl pyrophosphate isomerase (Idi), a farnesyl pyrophosphate synthetase (IspA), a geranyltranstransferase, an octoprenyl pyrophosphate synthase (IspB), a geranylgeranyl pyrophosphate (GGPP) synthase (CcrtE), a phytoene synthase (CrtB), a phytoene desaturase (CrtI), a lycopene cyclase (CrtY), a beta-carotene hydroxylase (CrtZ), a zeaxanthin glucosyl transferase (CrtX), a beta-carotene ketolase (CrtO), or their combination. Preferred Method: The genes in producing carotenoids are disrupted as mentioned in the bacterium cited above. The cell in enhanced production of carotenoids is contacted with one or more inhibitors of *gdhA* and *gpmB*, *gdhA* and *aceE*, *gdhA* and *ppc*, *gdhA* and *fdhF*, *gdhA* and *talB*, *fdhF*, *talB*, *gdhA*, *aceE* and *fdhF*, *gdhA* and *yjfp*, *gdhA*, *aceE* and *yjfp*, *gdhA*, *aceE*, *fdhF* and *yjfp*, *gpmB* and *yjiD*, *gdhA*, *aceE*, and *yjiD*, *gdhA*, *gpmB* and *yjiD*, *gdhA*, *aceE*, *hnr* and *yjfp*, *gdhA*, *aceE*, *hnr*, *yjfp* and *yjiD*, and/or *gdhA*, *gpmB* and *yjfp* expression or function. The one or more inhibitors comprise a nucleic acid. The cell is a bacterium. The method of enhancing production of carotenoids also comprises contacting a cell comprising genes involved in the carotenoid biosynthetic pathway with a plasmid comprising a *dxs*, *idi*, *yjiD*, *rpoS*, *torC*, *appY*, *ydgK*, *yeiA*, *yedR*, *tort*, *arcB*, *yggT*, *purDH*, *yfjN* gene, or their combination, culturing the cell where carotenoids are produced, and isolating carotenoids from the cell and enhancing production of carotenoids. The cell in any of the methods cited is a bacterium that belongs to the *Escherichia*, *Methylobacter*, *Methylococcus*, and *Methylosinus Salmonella*, *Erwina*, *Haematococcus*, *Rhodobacter*, *Myxococcus*, *Corynebacteria*, *Pseudomonas* or *Bacillus* genus.

The genes involved in the carotenoid biosynthetic pathway comprise an isopentenyl pyrophosphate isomerase (Idi), a farnesyl pyrophosphate synthetase (IspA), a geranyltranstransferase, an octoprenyl pyrophosphate synthase (IspB), a geranylgeranyl pyrophosphate (GGPP) synthase (CcrtE), a phytoene synthase (CrtB), a phytoene desaturase (CrtI), a lycopene cyclase (CrtY), a beta-carotene hydroxylase (CrtZ), a zeaxanthin glucosyl transferase (CrtX), a beta-carotene ketolase (CrtO), or their combination. Genetically disrupting *gdhA*, *gpmB*, *aceE*, *ppc*, *talB*, *fdhF*, *yjiD*, *hnr* or *yjfp* gene, or their combination results in an increased production of 3-95% as compared to wildtype cells. The method further comprises engineering the cell to over-express a *dxs*, *idi*, *yjiD*, *rpoS*, *torC*, *appY*, *ydgK*, *yeiA*, *yedR*, *tort*, *arcB*, *yggT*, *purDH*, or *yfjN* gene, or their combination. Engineering the cell to over-express the gene is accomplished with the use of a plasmid, which is a high- or low-copy plasmid. The carotenoids comprise *astaxanthin*, *canthaxanthin*, *beta-carotene*, *lycopene*, *phytoene* or *zeaxanthin*. The metabolite in determining optimized production of a metabolite comprises a product of the carotenoid biosynthetic pathway, and is *lycopene*. The *in silico* gene knockout simulations are conducted on one or more genes simultaneously or sequentially. The calculation of the flux profile comprises imposing a growth rate minimum that is equal to 5% of the maximum wild-type prediction of the flux balance analysis model. The metabolite in identifying genes involved in optimized production of a carotenoid

comprises a product of the carotenoid biosynthetic pathway, and is lycopene. The in silica gene knockout simulations are conducted on one or more genes simultaneously or sequentially. The calculation of the flux profile comprises imposing a growth rate minimum that is equal to 5% of the maximum wild-type prediction of the flux balance analysis model. The library of transposon mutagenized genes is produced using a pJA1 vector. The method further comprises enhancing gene expression of a gene involved in carotenoid biosynthesis in the cell. The expression of a *dxs*, *idi*, *yjiD*, *rpoS*, *torC*, *appY*, *ydgK*, *yeiA*, *yedR*, *tort*, *orcB*, *yggT*, *purDH*, *yjjN* or their combination, is enhanced. The cell is a bacterium, or is a yeast or mammalian. The metabolite in identifying genes involved in optimized production of a carotenoid comprises a product of the carotenoid biosynthetic pathway, and is lycopene. The in silica gene knockout simulations are conducted on one or more genes simultaneously or sequentially. The calculation of the flux profile comprises imposing a growth rate minimum that is equal to 5% of the maximum wild-type prediction of the flux balance analysis model. The library of transposon mutagenized genes is produced using a pJA1 vector. The method further comprises enhancing gene expression of a gene involved in carotenoid biosynthesis in the cell. The expression of a *dxs*, *idi*, *yjiD*, *rpoS*, *torC*, *appY*, *ydgK*, *yeiA*, *yedR*, *tort*, *orcB*, *yggT*, *purDH*, *yjjN* or their combination, is enhanced. The cell is a bacterium, or is a yeast or mammalian. Preferred Cell: The cell comprises a plasmid, which is a high- or low-copy plasmid. The gene is under the control of an inducible promoter. The cell is a bacterium that belongs to the *Escherichia*, *Methylobacter*, *Methylococcus*, and *Methylosinus Salmonella*, *Erwinia*, *Haematococcus*, *Rhodobacter*, *Myxococcus*, *Corynebacteria*, *Pseudomonas* or *Bacillus* genus. The cell further comprises a farnesyl pyrophosphate synthetase (*IspA*), a geranyltranstransferase, an octoprenyl pyrophosphate synthase (*IspB*), a geranylgeranyl pyrophosphate (GGPP) synthase (*CrtE*), a phytoene synthase (*CrtB*), a phytoene desaturase (*CrtI*), a lycopene cyclase (*CrtY*), a beta-carotene hydroxylase (*CrtZ*), a zeaxanthin glucosyl transferase (*CrtX*), a beta-carotene ketolase (*CrtO*), or their combination.

USE - The methods and compositions of the present invention are useful for overexpressing genes impacting, directly or indirectly, carotene biosynthesis, in particular for enhancing carotenoid production.

EXAMPLE - *Escherichia coli* K12 PT5-*dxs*, PT5-*idi*, PT5-*ispFD* was used as the lycopene expression strain when harboring the pAC-LYC plasmid containing the *crtEB1* operon. Gene deletions were conducted using PCR product recombination using the pKD46 plasmid expressing the lambda red recombination system and pKD13 as the kan template for PCR. (111 pages)

L8 ANSWER 2 OF 24 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN
DUPLICATE 2

ACCESSION NUMBER: 2005-06920 BIOTECHDS

TITLE: New nucleic acid molecule isolated from *Pantoea stewartii* strain DC413, encoding biosynthetic pathway enzyme, used for producing recombinant organisms capable of producing carotenoid compounds such as antheraxanthin, adonirubin and lycopene;
plasmid-mediated enzyme gene transfer and expression in bacterium, yeast or fungus for use in carotenoid production

AUTHOR: CHENG Q; TAO L; SEDKOVA N

PATENT ASSIGNEE: CHENG Q; TAO L; SEDKOVA N

PATENT INFO: US 2005014219 20 Jan 2005

APPLICATION INFO: US 2004-810733 26 Mar 2004

PRIORITY INFO: US 2004-810733 26 Mar 2004; US 2003-488183 17 Jul 2003

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2005-111250 [12]

AB DERWENT ABSTRACT:

NOVELTY - An isolated nucleic acid molecule (I) encoding a carotenoid biosynthetic pathway enzyme, is new.

DETAILED DESCRIPTION - An isolated nucleic acid molecule (I) encoding a carotenoid biosynthetic pathway enzyme, comprising an isolated nucleic acid molecule encoding a sequence of SEQ ID Nos 2, 4, 6, 8, 10, 12 and 14 of 302, 344, 429, 387, 493, 309 and 177 amino acids,

respectively fully defined in the specification, an isolated nucleic acid molecule that hybridizes with SEQ ID Nos 1, 3, 5, 7, 9, 11 and 13 of 909, 1035, 1290, 1164, 1482, 930 or 534 nucleotides long, respectively fully defined in the specification, and their complementary nucleic acid sequence, is new. INDEPENDENT CLAIMS are also included for: (1) a polypeptide (II) encoded by (I); (2) an isolated nucleic acid molecule (III) comprising a sequence (SEQ ID No 8) of 9127 nucleotides fully defined in the specification, comprising the crtE, idi, crtX, crtY, crtI, crtB and crtZ, genes are isolated genes or an isolated nucleic acid molecule having at least 95% identity to (SEQ ID No 8), where the isolated nucleic acid molecule encodes all of the polypeptides crtE, idi, crtX, crtY, crtI, crtB and crtZ; (3) an isolated nucleic acid molecule (IV) comprising a first nucleotide sequence encoding: (a) geranylgeranyl pyrophosphate synthetase enzyme of at least 302 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to the polypeptide having (SEQ ID No 2), (b) isopentenyl pyrophosphate isomerase enzyme of at least 344 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to the polypeptide having (SEQ ID No 4), (c) crtX enzyme of at least 429 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to the polypeptide having (SEQ ID No 6), (d) lycopene cyclase enzyme of at least 387 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to the polypeptide having (SEQ ID No 8), (e) phytoene desaturase enzyme of at least 493 amino acids that has at least 81% identity based on the Smith-Waterman method of alignment when compared to the polypeptide having (SEQ ID No 10), (f) phytoene synthase enzyme of at least 309 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to the polypeptide having (SEQ ID No 12), or (g) beta-carotene hydroxylase enzyme of at least 177 amino acids that has at least 82% identity based on the Smith-Waterman method of alignment when compared to the polypeptide having (SEQ ID No 14), or a second nucleotide sequence comprising the complement of the first nucleotide sequence; (4) a chimeric gene (V) comprising (I) or (IV) operably linked to suitable regulatory sequences; (5) a vector (VI) comprising (III); (6) a transformed host cell (VII) comprising (III) or (V); (7) regulating (M1) carotenoid biosynthesis in an organism, involves over-expressing at least one carotenoid biosynthetic pathway gene having (SEQ ID No 8) and a sequence selected from SEQ ID Nos 1, 3, 5, 7, 9, 11 and 13, in an organism such that the carotenoid biosynthesis is altered in the organism; (8) a strain DC413 comprising 16S rDNA sequence having a sequence (SEQ ID No 18) of 1351 nucleotides fully defined in the specification; (9) an isolated nucleic acid molecule encoding (SEQ ID Nos 2, 4, 6, 8, 10, 12 and 14); and (10) a method (M2) for producing carotenoid compounds, which involves providing host cell comprising a suitable levels of farnesyl pyrophosphate, and (I) or (III) under the control of suitable regulatory sequences, and contacting the host cell under suitable growth conditions with fermentable carbon substrate, thus producing a carotenoid compound.

BIOTECHNOLOGY - Preferred Nucleic Acid: (I) is chosen from (SEQ ID Nos 1, 3, 5, 7, 9, 11 and 13). (I) is isolated from *Pantoea stewartii* strain DC413. Preferred Host Cell: (VI) is chosen from bacteria, yeast, filamentous fungi, algae and green plants, preferably *Aspergillus*, *Trichoderma*, *Saccharomyces*, *Pichia*, *Candida*, *Hansenula*, *Yarrowia*, *Rhodospiridium*, *Lipomyces*, *Salmonella*, *Bacillus*, *Acinetobacter*, *Zymomonas*, *Agrobacterium*, *Flavobacterium*, *Rhodobacter*, *Rhodococcus*, *Streptomyces*, *Brevibacterium*, *Corynebacteria*, *Mycobacterium*, *Escherichia*, *Pantoea*, *Pseudomonas*, *Methylobacter*, *Methylococcus*, *Methylosinus*, *Methylomicrobium*, *Methylocystis*, *Alcaligenes*, *Synechocystis*, *Synechococcus*, *Anabaena*, *Thiobacillus*, *Methanobacterium*, *Klebsiella*, *Methylophilus*, *Methylobacillus*, *Methylobacterium*, *Hyphomicrobium*, *Xanthobacter*, *Paracoccus*, *Norcardia*, *Arthrobacter*, *Rhodopseudomonas*, *Torulopsis*, *Phaffia*, and *Rhodotorula*. Preferred Method: In (M1), the carotenoid gene is over-expressed on a multicopy plasmid. The carotenoid gene is operably linked to an inducible or regulated promoter. The carotenoid gene is expressed in antisense orientation. The carotenoid gene is disrupted by insertion of foreign DNA into the coding region. In (M2), the transformed host cell is chosen from 1C metabolizing

hosts, bacteria, yeast, filamentous fungi, algae and green plants. The 1C metabolizing host is a methanoptroph and the fermentable carbon substrate is chosen from methane, methanol, formaldehyde, formic acid, methylated amines, methylated thiols and carbondioxide. The 1C metabolizing host grows on 1C carbon substrate chosen from methane and methanol, and comprises a functional Embden-Meyerhof carbon pathway, the pathway comprising a gene encoding a pyrophosphate-dependent phosphofructokinase enzyme. The 1C metabolizing host cell is a high growth methanotrophic bacterial strain, known as Methylomonas 16a (ATCC designation PTA 2402).

USE - (I) is useful for producing carotenoid compounds. (I) is useful for producing recombinant organisms that have the ability to produce carotenoid compounds.

EXAMPLE - To isolate carotenoid-producing bacterial strains, pigmented microbes were isolated from a collection of environmental samples. A soil sample from Florida was collected and resuspended in Luria-Broth (LB). A 10 microlitres loopful of cell suspension was streaked onto LB plates and the plates were incubated at 30 degreesC. Pigmented bacteria with diverse colony appearances were picked and streaked twice to homogeneity on LB plates and incubated at 30 degreesC. From these colonies, one which formed shiny yellow colonies was designated as strain DC413. The yellow pigment in Pantoea stewartii DC413 was extracted and analyzed by HPLC. The strain was grown in 100 ml LB at 30 degreesC for 2 days and then cells were harvested by centrifugation at 4000 g for 30 minutes. The cell pellet was extracted with 10 ml acetone. The solvent was dried under nitrogen and the carotenoids were resuspended in 0.5 ml acetone. The extraction was filtered with an Acrodisc CR25 mm syringe filter and then concentrated in 0.1 ml 10% acetone+90% acetonitrile for HPLC analysis. sample (20 microlitres) was loaded onto ZORBAX C18 column. HPLC analysis indicated that strain DC413 produced zeaxanthin (6.27 minutes peak) and beta-carotene (13.01 minutes peak) by comparison with authentic standards of zeaxanthin. MS analysis confirmed that the molecular weight of the zeaxanthin peak was 569, and that of the beta-carotene peak was 537. The predominant peak that eluted at 3.24 minutes was most likely zeaxanthin monoglucoside, as suggested by its molecular weight of 731. (51 pages)

L8 ANSWER 3 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STM

ACCESSION NUMBER: 2005:182845 HCAPLUS

DOCUMENT NUMBER: 142:275033

TITLE: Method for producing ketocarotenoids in genetically modified, non-human organisms

INVENTOR(S): Flachmann, Ralf; Schopfer, Christel Renate; Herbers, Karin; Kunze, Irene; Sauer, Matt; Klebsattel, Martin; Luck, Thomas; Voeste, Dirk; Pfeiffer, Angelika-Maria

PATENT ASSIGNEE(S): Sungene GmbH & Co. Kgaa, Germany

SOURCE: PCT Int. Appl., 358 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: German

FAMILY ACC. NUM. COUNT: 14

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2005019467	A1	20050303	WO 2004-EP8623	20040731
W:	AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW			
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WO 2004018693	A2	20040304	WO 2003-EP309102	20030818
WO 2004018693	A3	20041209		
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WO 2004018385 A2 20040304 WO 2003-EP309105 20030818

WO 2004018385 A3 20041021

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WO 2004018695 A2 20040304 WO 2003-EP309107 20030818

WO 2004018695 A3 20041014

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN,
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DE 102004007622 A1 20050825 DE 2004-102004007622 20040217

PRIORITY APPLN. INFO.:

EP 2003-9101 A 20030818
 EP 2003-9106 A 20030818
 EP 2003-9109 A 20030818
 WO 2003-EP9102 A 20030818
 WO 2003-EP9105 A 20030818
 WO 2003-EP9107 A 20030818
 DE 2004-102004007622A 20040217
 DE 2002-10238978 A 20020820
 DE 2002-10238979 A 20020820
 DE 2002-10238980 A 20020820
 DE 2002-10253112 A 20021113
 DE 2002-10258971 A 20021216

AB The invention relates to a method for producing ketocarotinoids by
 cultivation of genetically modified organisms that have a modified
 ketolase activity and modified β -cyclase activity as compared to the
 wild-type organism. The invention also relates to the genetically
 modified organisms, to their use as food or feed, and to their use for
 producing ketocarotenoid exts. Thus, transgenic tomato and marigold
 plants expressing Nostoc punctiforme ketolase cDNA were created.

REFERENCE COUNT: 8 THERE ARE 8 CITED REFERENCES AVAILABLE FOR THIS
 RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 4 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2005:182839 HCAPLUS

DOCUMENT NUMBER: 142:276001

TITLE: Novel ketolases and the genes encoding them and their
 use in the manufacture of carotenoids with transgenic
 organisms

INVENTOR(S): Sauer, Matt; Schopfer, Christel Renate; Flachmann,
 Ralf; Herbers, Karin; Kunze, Irene; Klebsattel,
 Martin; Luck, Thomas; Voeste, Dirk; Pfeiffer,
 Angelika-Maria; Tschoep, Hendrik

PATENT ASSIGNEE(S): Sungene GmbH & Co. Kgaa, Germany

SOURCE: PCT Int. Appl., 317 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent
 LANGUAGE: German
 FAMILY ACC. NUM. COUNT: 14
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2005019461	A2	20050303	WO 2004-EP8625	20040731
WO 2005019461	A3	20050616		
W:	AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW			
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WO 2004018688	A1	20040304	WO 2003-EP309101	20030818
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WO 2004018693	A2	20040304	WO 2003-EP309102	20030818
WO 2004018693	A3	20041209		
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WO 2004018385	A2	20040304	WO 2003-EP309105	20030818
WO 2004018385	A3	20041021		
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WO 2004018694	A2	20040304	WO 2003-EP309106	20030818
WO 2004018694	A3	20040910		
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WO 2004018695	A2	20040304	WO 2003-EP309107	20030818
WO 2004018695	A3	20041014		

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WO 2004017749 A2 20040304 WO 2003-EP309109 20030818

WO 2004017749 A3 20041014

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DE 102004007624 A1 20050915 DE 2004-102004007624 20040217

PRIORITY APPLN. INFO.:

WO 2003-EP9101 A 20030818
 WO 2003-EP9102 A 20030818
 WO 2003-EP9105 A 20030818
 WO 2003-EP9106 A 20030818
 WO 2003-EP9107 A 20030818
 WO 2003-EP9109 A 20030818
 DE 2004-102004007624A 20040217
 DE 2002-10238978 A 20020820
 DE 2002-10238979 A 20020820
 DE 2002-10238980 A 20020820
 DE 2002-10253112 A 20021113
 DE 2002-10258971 A 20021216

AB The invention relates to a method for producing ketocarotenoids in flowers by cultivation of genetically modified, non-human organisms expressing a foreign ketolase gene. Novel ketolases that play a key role in carotenoid biosynthesis are identified in algae and cyanobacteria. Expression of these genes, in combination with genes for other enzymes of the carotenoid biosynthetic pathway may be used to improve the flux through the pathway and overall yield of carotenoids. The invention also relates to the genetically modified organisms, their use as food stuff and feeding stuff and to their use for producing ketocarotenoid exts. and to novel ketolases and nucleic acids encoding said ketolases.

L8 ANSWER 5 OF 24

MEDLINE on STN

DUPLICATE 3

ACCESSION NUMBER: 2005030607 MEDLINE

DOCUMENT NUMBER: PubMed ID: 15657364

TITLE: Carotenoids activate the antioxidant response element transcription system.

AUTHOR: Ben-Dor Anat; Steiner Michael; Gheber Larisa; Danilenko Michael; Dubi Noga; Linnewiel Karin; Zick Anat; Sharoni Yoav; Levy Joseph

CORPORATE SOURCE: Department of Clinical Biochemistry, Faculty of Health Sciences, Ben-Gurion University of the Negev, Beer-Sheva, Israel.

SOURCE: Molecular cancer therapeutics, (2005 Jan) 4 (1) 177-86. Journal code: 101132535. ISSN: 1535-7163.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200505

ENTRY DATE: Entered STN: 20050120

Last Updated on STN: 20050513

Entered Medline: 20050512

AB Epidemiologic studies have found an inverse association between consumption of tomato products and the risk of certain types of cancers.

However, the mechanisms underlying this relationship are not completely understood. One mechanism that has been suggested is induction of phase II detoxification enzymes. Expression of phase II enzymes is regulated by the antioxidant response element (ARE) and the transcription factor Nrf2 (nuclear factor E2-related factor 2). In this study, we determined the role of this transcription system in the induction of phase II enzymes by carotenoids. We found that in transiently transfected cancer cells, lycopene transactivated the expression of reporter genes fused with ARE sequences. Other carotenoids such as phytoene, phytofluene, beta-carotene, and astaxanthin had a much smaller effect. An increase in protein as well as mRNA levels of the phase II enzymes NAD(P)H:quinone oxidoreductase and gamma-glutamylcysteine synthetase was observed in nontransfected cells after carotenoid treatment. Ethanolic extract of lycopene containing unidentified hydrophilic derivatives of the carotenoid activated ARE with similar potency to lycopene. The potency of the carotenoids in ARE activation did not correlate with their effect on intracellular reactive oxygen species and reduced glutathione level, which may indicate that ARE activation is not solely related to their antioxidant activity. Nrf2, which is found predominantly in the cytoplasm of control cells, translocated to the nucleus after carotenoid treatment. Interestingly, part of the translocated Nrf2 colocalized with the promyelocytic leukemia protein in the promyelocytic leukemia nuclear bodies. The increase in phase II enzymes was abolished by a dominant-negative Nrf2, suggesting that carotenoid induction of these proteins depends on a functional Nrf2 and the ARE transcription system.

L8 ANSWER 6 OF 24 EMBASE COPYRIGHT (c) 2005 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 2005121198 EMBASE
 TITLE: Diversifying carotenoid biosynthetic pathways by directed evolution.
 AUTHOR: Umeno D.; Tobias A.V.; Arnold F.H.
 CORPORATE SOURCE: F.H. Arnold, Div. of Chem. and Chem. Engineering, CA Institute of Technology, 210-41, 1200 E. California Blvd., Pasadena, CA 91125, United States. frances@cheme.caltech.edu
 SOURCE: Microbiology and Molecular Biology Reviews, (2005) Vol. 69, No. 1, pp. 51-78. Refs: 240 ISSN: 1092-2172 CODEN: MMBRF7
 COUNTRY: United States
 DOCUMENT TYPE: Journal; General Review
 FILE SEGMENT: 004 Microbiology 029 Clinical Biochemistry
 LANGUAGE: English
 SUMMARY LANGUAGE: English
 ENTRY DATE: Entered STN: 20050331 Last Updated on STN: 20050331

AB Microorganisms and plants synthesize a diverse array of natural products, many of which have proven indispensable to human health and well-being. Although many thousands of these have been characterized, the space of possible natural products - those that could be made biosynthetically - remains largely unexplored. For decades, this space has largely been the domain of chemists, who have synthesized scores of natural product analogs and have found many with improved or novel functions. New natural products have also been made in recombinant organisms, via engineered biosynthetic pathways. Recently, methods inspired by natural evolution have begun to be applied to the search for new natural products. These methods force pathways to evolve in convenient laboratory organisms, where the products of new pathways can be identified and characterized in high-throughput screening programs. Carotenoid biosynthetic pathways have served as a convenient experimental system with which to demonstrate these ideas. Researchers have mixed, matched, and mutated carotenoid biosynthetic enzymes and screened libraries of these "evolved" pathways for the emergence of new carotenoid products. This has led to dozens of new pathway products not previously known to be made by the assembled enzymes. These new products include whole families of carotenoids built from backbones not found in nature. This review details the strategies

and specific methods that have been employed to generate new carotenoid biosynthetic pathways in the laboratory. The potential application of laboratory evolution to other biosynthetic pathways is also discussed. Copyright .COPYRG. 2005, American Society for Microbiology. All Rights Reserved.

L8 ANSWER 7 OF 24 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN
DUPLICATE 4

ACCESSION NUMBER: 2005-01718 BIOTECHDS

TITLE: New nucleic acid molecules isolated from strain DC260 and encoding a carotenoid biosynthetic enzyme, useful for producing carotenoid compounds, and for regulating carotenoid biosynthesis;
for use in carotenoid synthesis and regulation

AUTHOR: CHENG Q; TAO L

PATENT ASSIGNEE: DU PONT DE NEMOURS and CO E I

PATENT INFO: WO 2004104180 2 Dec 2004

APPLICATION INFO: WO 2004-US15524 14 May 2004

PRIORITY INFO: US 2003-527083 3 Dec 2003; US 2003-471904 20 May 2003

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2005-013283 [01]

AB DERWENT ABSTRACT:

NOVELTY - An isolated nucleic acid molecule encoding a carotenoid biosynthetic enzyme, is new.

DETAILED DESCRIPTION - The isolated nucleic acid molecule is selected from: (a) an isolated nucleic acid molecule encoding the amino acid sequence of 301, 425, 388, 493, 309, or 177 (P1-P6) amino acids, fully defined in the specification; (b) an isolated nucleic acid molecule that hybridizes with (a) under the following hybridization conditions: 0.1 x SSC, 0.1% SDS, 65 degrees C and washed with 2 x SSC, 0.1% SDS followed by 0.1 x SSC, 0.1% SDS; or (c) an isolated nucleic acid molecule that is complementary to (a) or (b). INDEPENDENT CLAIMS are included for: (1) an isolated nucleic acid fragment of the nucleic acid cited above isolated from strain DC260; (2) a polypeptide encoded by the isolated nucleic acid molecule; (3) an isolated nucleic acid molecule of 6999 bp (S1), fully defined in the specification, comprising the crtE, crtX, crtY, crtI, crtB and crtZ, genes or an isolated nucleic acid molecule having at least 95% identity to S1; (4) an isolated nucleic acid molecule comprising a first nucleotide sequence encoding a geranylgeranyl pyrophosphate synthetase enzyme of at least 301 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to a polypeptide having the sequence pf P1, or a second nucleotide sequence comprising the complement of the first nucleotide sequence; (5) an isolated nucleic acid molecule comprising a first nucleotide sequence encoding a zeaxanthin glucosyl transferase enzyme of at least 425 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to a polypeptide having the sequence of P2, or a second nucleotide sequence comprising the complement of the first nucleotide sequence; (6) an isolated nucleic acid molecule comprising a first nucleotide sequence encoding a lycopene cyclase enzyme of at least 388 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to a polypeptide having the sequence as set forth P3, or a second nucleotide sequence comprising the complement of the first nucleotide sequence; (7) an isolated nucleic acid molecule comprising a first nucleotide sequence encoding a phytoene desaturase enzyme of at least 493 amino acids that has at least 77% identity based on the Smith-Waterman method of alignment when compared to a polypeptide having the sequence of P4, or a second nucleotide sequence comprising the complement of the first nucleotide sequence; (8) an isolated nucleic acid molecule comprising a first nucleotide sequence encoding a phytoene synthase enzyme of at least 309 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to a polypeptide having the sequence of P5, or a second nucleotide sequence comprising the complement of the first nucleotide sequence; (9) an isolated nucleic acid molecule comprising a first nucleotide sequence encoding a beta-carotene hydroxylase enzyme of at least 177 amino acids that has at least 70% identity based on the Smith-Waterman method of alignment when compared to

a polypeptide having the sequence of P6, or a second nucleotide sequence comprising the complement of the first nucleotide sequence; (10) a chimeric gene comprising the isolated nucleic acid molecule operably linked to suitable regulatory sequences; (11) a vector comprising the isolated nucleic acid molecule; (12) a transformed host cell comprising the chimeric gene; (13) a transformed host comprising the isolated nucleic acid molecule; (14) producing carotenoid compounds; (15) regulating carotenoid biosynthesis in an organism; (16) a strain DC260 comprising the 16s rDNA sequence of 1327 bp, fully defined in the specification.

BIOTECHNOLOGY - Preferred Cell: The transformed host cell is selected from bacteria, yeast, filamentous fungi, algae, and green plants. The host cell is selected from *Aspergillus*, *Trichoderma*, *Saccharomyces*, *Pichia*, *Candida*, *Hansenula*, *Yarrowia*, *Rhodospiridium*, *Lipomyces*, *Salmonella*, *Bacillus*, *Acinetobacter*, *Zymomonas*, *Agrobacterium*, *Flavobacterium*, *Rhodobacter*, *Rhodococcus*, *Streptomyces*, *Brevibacterium*, *Corynebacteria*, *Mycobacterium*, *Escherichia*, *Pantoea*, *Pseudomonas*, *Methylomonas*, *Methylobacter*, *Methylococcus*, *Methylosinus*, *Methylobacterium*, *Methylocystis*, *Alcaligenes*, *Synechocystis*, *Synechococcus*, *Anabaena*, *Thiobacillus*, *Methanobacterium*, *Klebsiella*, *Methylophilus*, *Methylobacillus*, *Methylobacterium*, *Hyphomicrobium*, *Xanthobacter*, *Paracoccus*, *Nocardia*, *Arthrobacter*, *Rhodopseudomonas*, *Torulopsis*, *Phaffia*, and *Rhodotorula*. Preferred Method: Producing carotenoid compounds comprises providing a transformed host cell comprising suitable levels of farnesyl pyrophosphate, and a set of nucleic acid molecules encoding the enzymes of P1-P6 under the control of suitable regulatory sequences; and contacting the host cell under suitable growth conditions with an effective amount of a fermentable carbon substrate whereby a carotenoid compound is produced. The transformed host cell is selected from C1 metabolizing hosts, bacteria, yeast, filamentous fungi, algae, and green plants. The C1 metabolizing host is a methanotroph and the fermentable carbon substrate is selected from methane, methanol, formaldehyde, formic acid, methylated amines, methylated thiols, and carbon dioxide. The C1 metabolizing host grows on a C1 carbon substrate selected from methane and methanol; and comprises a functional Embden-Meyerhof carbon pathway, the pathway comprising a gene encoding a pyrophosphate-dependent phosphofructokinase enzyme. The C1 metabolizing host cell is a high growth methanotrophic bacterial strain, known as *Methylomonas* 16a and having the American Type Culture Collection ATCC designated as PTA 2402. The carotenoid compound produced is selected from antheraxanthin, adonirubin, adonixanthin, **astaxanthin**, canthaxanthin, capsorubin, beta-cryptoxanthin, alpha-carotene, beta-carotene, epsilon-carotene, echinenone, 3-hydroxyechinenone, 3'-hydroxyechinenone, gamma-carotene, 4-keto-gamma-carotene, alpha-cryptoxanthin, deoxyflexixanthin, diatoxanthin, 7,8-didehydroastaxanthin, fucoxanthin, fucoxanthinol, isorenieratene, lactucaxanthin, lutein, lycopene, myxobactone, neoxanthin, neurosporene, hydroxyneurosporene, peridinin, phytoene, rhodopin, rhodopin glucoside, 4-keto-rubixanthin, siphonaxanthin, spheroidene, spheroidenone, spirilloxanthin, 4-keto-torulene, 3-hydroxy-4-keto-torulene, uriolide, uriolide acetate, violaxanthin, zeaxanthin-beta-diglucoside, and zeaxanthin. Regulating carotenoid biosynthesis in an organism comprises over-expressing at least one carotenoid gene in an organism such that the carotenoid biosynthesis is altered in the organism. The carotenoid gene is over-expressed on a multicopy plasmid. The carotenoid gene is operably linked to an inducible or regulated promoter. The carotenoid gene is expressed in antisense orientation. The carotenoid gene is disrupted by insertion of foreign DNA into the coding region.

USE - The nucleic acid molecules are useful for producing carotenoid compounds (claimed) by converting farnesyl pyrophosphates to carotenoids, and for regulating carotenoid biosynthesis (claimed).

EXAMPLE - Genes encoding carotenoid biosynthetic enzyme, e.g. geranylgeranyl pyrophosphate, **synthetase**, phytoene synthase, phytoene desaturase, lycopene cyclase, beta-carotene hydroxylase, and zeaxanthin glucosyl transferase were identified by conducting Basic Local Alignment Search Tool (BLAST) searches for similarity to sequences contained in the BLAST nr database. The sequence was analyzed for similarity to all publicly available DNA sequences contained in the database using the BLASTN algorithm. The DNA sequence was translated in

all reading frames and compared for similarity to all publicly available protein sequences. (95 pages)

L8 ANSWER 8 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2004:546564 HCAPLUS
DOCUMENT NUMBER: 141:105355
TITLE: Mutant carotenoid-producing microorganisms and their use in carotenoid fermentation
INVENTOR(S): Cheng, Qiong; Rouviere, Pierre E.; Tao, Luan
PATENT ASSIGNEE(S): E.I. Du Pont De Nemours and Company, USA
SOURCE: PCT Int. Appl., 85 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004056974	A2	20040708	WO 2003-US41811	20031219
W: AU, CA, JP, NO				
RW: AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR				
US 2004146966	A1	20040729	US 2003-735008	20031212
CA 2509866	AA	20040708	CA 2003-2509866	20031219
EP 1581632	A2	20051005	EP 2003-800443	20031219
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, FI, RO, CY, TR, BG, CZ, EE, HU, SK				
PRIORITY APPLN. INFO.:			US 2002-435612P	P 20021219
			WO 2003-US41811	W 20031219

AB Disclosed are carotenoid-overproducing microorganisms (yeast, filamentous fungi, bacteria) with disrupted deadD, mreC, and/or yfhE genes and fermentative manufacture of carotenoids with cultures of these microorganisms. Thus, mutations in genes having no direct relationship to the carotenoid biosynthetic pathway have been found to increase carbon flux through that pathway. Complete disruption in the deadD, mreC, and yfhE genes of E. coli were effective. Addnl. where genes of the lower carotenoid pathway (i.e., crtEXYIB genes) reside on a plasmid having either a p15A or pMB1 replicon, mutations in the thrS, rspA, rpoC, yjeR, and rhoL were found effective.

L8 ANSWER 9 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2004:183041 HCAPLUS
DOCUMENT NUMBER: 140:234476
TITLE: Transgenic plants with enhanced ketolase activity for use in production of ketocarotenoids in flower petals
INVENTOR(S): Schopfer, Christel Renate; Flachmann, Ralf; Herbers, Karin; Kunze, Irene; Sauer, Matt; Klebsattel, Martin
PATENT ASSIGNEE(S): Sungene G.m.b.H. & Co. K.-G.a.A., Germany
SOURCE: PCT Int. Appl., 497 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: German
FAMILY ACC. NUM. COUNT: 14
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2004018693	A2	20040304	WO 2003-EP309102	20030818
WO 2004018693	A3	20041209		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

DE 10238979	A1	20040226	DE 2002-10238979	20020820
DE 10238978	A1	20040304	DE 2002-10238978	20020820
DE 10238980	A1	20040304	DE 2002-10238980	20020820
DE 10253112	A1	20040603	DE 2002-10253112	20021113
DE 10258971	A1	20040701	DE 2002-10258971	20021216
CA 2496133	AA	20040304	CA 2003-2496133	20030818
EP 1532264	A2	20050525	EP 2003-792345	20030818
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK				
WO 2005019467	A1	20050303	WO 2004-EP8623	20040731
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
WO 2005019460	A2	20050303	WO 2004-EP8624	20040731
WO 2005019460	A3	20050721		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				
WO 2005019461	A2	20050303	WO 2004-EP8625	20040731
WO 2005019461	A3	20050616		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW				
RW: BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG				

PRIORITY APPLN. INFO.:

DE 2002-10238978	A	20020820
DE 2002-10238979	A	20020820
DE 2002-10238980	A	20020820
DE 2002-10253112	A	20021113
DE 2002-10258971	A	20021216
EP 2003-9101	A	20030818
EP 2003-9106	A	20030818
EP 2003-9109	A	20030818
WO 2003-EP9101	A	20030818
WO 2003-EP9102	W	20030818
WO 2003-EP9105	A	20030818
WO 2003-EP9106	A	20030818
WO 2003-EP9107	A	20030818
WO 2003-EP9109	A	20030818
DE 2004-102004007622A		20040217
DE 2004-102004007623A		20040217
DE 2004-102004007624A		20040217

AB The invention relates to a method for the production of ketocarotinoids by means of the cultivation of plants, which have an altered ketolase activity in flower petals in comparison to the wild type, the genetically altered plants and the use thereof as human and animal foodstuffs and for the production of ketocarotinoid exts. Thus, the flower petals of transgenic

tomato expressing the *Haematococcus pluvialis* ketolase gene from the flower-specific AP3 promoter of *A. thaliana* were examined for ketocarotenoid content. While control plants produced primarily violaxanthin, the transgenic plants produced mainly **astaxanthin** (61% of total carotenoids) with some adonirubin (15%), adonixanthin (4%), and β/ζ -carotene (15%), and no violaxanthin. Similar results were observed with transgenic *Tagetes erecta*.

L8 ANSWER 10 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN
 ACCESSION NUMBER: 2004:1081995 HCAPLUS
 DOCUMENT NUMBER: 142:50293
 TITLE: Gene cluster encoding carotenoid biosynthetic enzymes from *Pantoea agglomerans*
 INVENTOR(S): Cheng, Qiong; Sedkova, Natalia; Tao, Luan
 PATENT ASSIGNEE(S): E. I. Du Pont De Nemours and Company, USA
 SOURCE: U.S. Pat. Appl. Publ., 49 pp.
 CODEN: USXXCO
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 5
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2004253663	A1	20041216	US 2004-808807	20040324
US 6929928	B2	20050816		
WO 2005044975	A2	20050519	WO 2004-US19038	20040610

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BW, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NA, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW

RW: BW, GH, GM, KE, LS, MW, MZ, NA, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

PRIORITY APPLN. INFO.: US 2003-477874P P 20030612
 US 2003-527083P P 20031203

AB A unique carotenogenic biosynthetic gene cluster was isolated from *Pantoea agglomerans* strain DC404, wherein the genetic organization of the cluster is crtE-idi-crtY-crtI-crtB-crtZ. The genes contained within this cluster encode geranylgeranyl pyrophosphate (GGPP) **synthetase** (CrtE), isopentenyl pyrophosphate isomerase (Idi), lycopene cyclase (CrtY), phytoene desaturase (CrtI), phytoene synthase (CrtB), and β -carotene hydroxylase (CrtZ). The gene cluster, genes and their products are useful for the conversion of farnesyl pyrophosphate to carotenoids. Vectors containing those DNA segments, host cells containing the vectors, and methods for producing those enzymes by recombinant DNA technol. in transformed host organisms are disclosed.

L8 ANSWER 11 OF 24 EMBASE COPYRIGHT (c) 2005 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 2004123707 EMBASE
 TITLE: Cell wall proteomics of the green alga *Haematococcus pluvialis* (Chlorophyceae).
 AUTHOR: Wang S.-B.; Hu Q.; Sommerfeld M.; Chen F.
 CORPORATE SOURCE: Q. Hu, School of Life Sciences, Arizona State University, P.O. Box 874501, Tempe, AZ 85287-4501, United States. huqiang@asu.edu
 SOURCE: Proteomics, (2004) Vol. 4, No. 3, pp. 692-708.
 Refs: 64
 ISSN: 1615-9853 CODEN: PROTC7
 COUNTRY: Germany
 DOCUMENT TYPE: Journal; Article
 FILE SEGMENT: 004 Microbiology
 LANGUAGE: English
 SUMMARY LANGUAGE: English

ENTRY DATE: Entered STN: 20040412
Last Updated on STN: 20040412

AB The green microalga *Haematococcus pluvialis* can synthesize and accumulate large amounts of the ketocarotenoid **astaxanthin**, and undergo profound changes in cell wall composition and architecture during the cell cycle and in response to environmental stresses. In this study, cell wall proteins (CWPs) of *H. pluvialis* were systematically analyzed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) coupled with peptide mass fingerprinting (PMF) and sequence-database analysis. In total, 163 protein bands were analyzed, which resulted in positive identification of 81 protein orthologues. The highly complex and dynamic composition of CWPs is manifested by the fact that the majority of identified CWPs are differentially expressed at specific stages of the cell cycle along with a number of common wall-associated 'housekeeping' proteins. The detection of cellulose synthase orthologue in the vegetative cells suggested that the biosynthesis of cellulose occurred during primary wall formation, in contrast to earlier observations that cellulose was exclusively present in the secondary wall of the organism. A transient accumulation of a putative cytokinin oxidase at the early stage of encystment pointed to a possible role in cytokinin degradation while facilitating secondary wall formation and/or assisting in cell expansion. This work represents the first attempt to use a proteomic approach to investigate CWPs of microalgae. The reference protein map constructed and the specific protein markers obtained from this study provide a framework for future characterization of the expression and physiological functions of the proteins involved in the biogenesis and modifications in the cell wall of *Haematococcus* and related organisms.

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ACCESSION NUMBER: 2004260833 EMBASE
TITLE: Study of the expression of carotenoid biosynthesis genes in wild-type and deregulated strains of *xanthophyllomyces dendrorhous* (ex: *Phaffia rhodozyma*).
AUTHOR: Lodato P.; Alcaino J.; Barahona S.; Retamales P.; Jimenez A.; Cifuentes V.
CORPORATE SOURCE: V. Cifuentes, Casilla 653, Santiago, Chile.
vcifuent@uchile.cl
SOURCE: Biological Research, (2004) Vol. 37, No. 1, pp. 83-93.
Refs: 44
ISSN: 0716-9760 CODEN: BESEEB
COUNTRY: Chile
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 20040709
Last Updated on STN: 20040709

AB The expression, at the mRNA level, of carotenoid biosynthetic genes from *Xanthophyllomyces dendrorhous* was studied by RT-PCR. The experimental conditions for the RT-PCR assay were standardized to quantify the relative transcript levels of *idi*, *crtE*, *crtYB* and *crtI* genes. This work attempted to correlate **astaxanthin** production with the transcript levels of carotenogenic genes in a wild-type strain (UCD 67-385) and two overproducer deregulated strains (*atxS1* and *atxS2*). At 3 day cultures, the wild-type strain contained higher transcript levels from the *crtE* and *crtYB* genes on minimal medium than on rich medium. Similarly, carotenoid production was higher on minimal medium than on rich medium. However, carotenoid production in the *atxS1* and *atxS2* strains was not correlated with the transcript level of carotenogenic genes under the same experimental conditions. This result suggests that there is not a linear relationship between carotenogenic transcript levels and carotenoid biosynthesis.

L8 ANSWER 13 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2003:355739 HCAPLUS
DOCUMENT NUMBER: 138:363828
TITLE: Isolated carotenoid biosynthesis gene cluster from *Bradyrhizobium* involved in canthaxanthin production

use as a feed additive
 INVENTOR(S): Giraud, Eric; Hannibal, Laure
 PATENT ASSIGNEE(S): Fr.
 SOURCE: U.S. Pat. Appl. Publ., 14 pp.
 CODEN: USXXCO
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 2003087337	A1	20030508	US 2002-166037	20020611
CA 2349040	AA	20021213	CA 2001-2349040	20010613
PRIORITY APPLN. INFO.:			US 2001-297272P	P 20010612

AB Isolated gene cluster involved in canthaxanthin biosynthesis, which comprises a polynucleotide wherein: crtY, crtI, crtB and crtW genes are clustered in this order and in the same orientation, and, preceding the four cited genes, crtE gene is oriented in the opposite direction. Applications for producing natural carotenoids useful in pharmaceutical, cosmetic and nutritious compns.

L8 ANSWER 14 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2003:922684 HCAPLUS
 DOCUMENT NUMBER: 140:1581
 TITLE: Regulation of genes involved in carotenoid and tocopherol biosynthesis pathway in transgenic plants for producing carotenoid compounds, tocopherol compounds, and specialty oils in plant seeds
 INVENTOR(S): Shewmaker, Christine K.; Bhat, B. Ganesh; Venkatramesh, Mylavaraapu; Rangwala, Shaikat H.; Kishore, Ganesh M.; Boddupalli, Sekhar S.
 PATENT ASSIGNEE(S): Calgene LLC, USA
 SOURCE: U.S., 57 pp.
 CODEN: USXXAM
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6653530	B1	20031125	US 1998-23587	19980213
PRIORITY APPLN. INFO.:			US 1998-23587	19980213

AB Methods are provided for producing plants and seeds having altered carotenoid, fatty acid and tocopherol compns. The methods find particular use in increasing the carotenoid and tocopherol levels in oilseed plants, and in providing desirable high oleic acid seed oils. Specifically, chimeric genes encoding E. ureidovora phytoene synthase (crtB), or phytoene desaturase (crtI), or GGPP synthase (crtE) in fusion with plastid transit peptide of pea Rubisco small subunit (rbcS) under the control of seed-preferred napin gene promoter, and napin or nos termination region, are constructed to make transgenic Brassica napus. Binary constructs expressing both crtB and crtI genes, crtB and antisense epsilon cyclase or beta cyclase genes are also used to transform Brassica napus. Also demonstrated are increased carotenoid production, particularly increased ratio of α -carotene and β -carotene to phytoene, and increased levels of oleic acid and decreased levels of linoleic and/or linolenic acid in seeds of transgenic plants.

REFERENCE COUNT: 104 THERE ARE 104 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 15 OF 24 EMBASE COPYRIGHT (c) 2005 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 2003291923 EMBASE
 TITLE: Metabolic engineering of the carotenoid biosynthetic pathway in the yeast Xanthophyllomyces dendrorhous (Phaffia rhodozyma).

AUTHOR: Verdoes J.C.; Sandmann G.; Visser H.; Diaz M.; Van Mossel M.; Van Ooyen A.J.J.
CORPORATE SOURCE: H. Visser, Laboratory of Microbiology, Dept. of Agrotechnology/Food Sci., Wageningen University, Dreijenlaan 2, 6703 HA Wageningen, Netherlands. hans.visser@wur.nl
SOURCE: Applied and Environmental Microbiology, (1 Jul 2003) Vol. 69, No. 7, pp. 3728-3738.
Refs: 36
ISSN: 0099-2240 CODEN: AEMIDF
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 20030810
Last Updated on STN: 20030810

AB The crtYB locus was used as an integrative platform for the construction of specific carotenoid biosynthetic mutants in the astaxanthin-producing yeast *Xanthophyllomyces dendrorhous*. The crtYB gene of *X. dendrorhous*, encoding a chimeric carotenoid biosynthetic enzyme, could be inactivated by both single and double crossover events, resulting in non-carotenoid-producing transformants. In addition, the crtYB gene, linked to either its homologous or a glyceraldehyde-3-phosphate dehydrogenase promoter, was overexpressed in the wild type and a β -carotene-accumulating mutant of *X. dendrorhous*. In several transformants containing multiple copies of the crtYB gene, the total carotenoid content was higher than in the control strain. This increase was mainly due to an increase of the β -carotene and echinone content, whereas the total content of astaxanthin was unaffected or even lower. Overexpression of the phytoene synthase-encoding gene (crtI) had a large impact on the ratio between mono- and bicyclic carotenoids. Furthermore, we showed that in metabolic engineered *X. dendrorhous* strains, the competition between the enzymes phytoene desaturase and lycopene cyclase for lycopene governs the metabolic flux either via β -carotene to astaxanthin or via 3,4-didehydrolycopene to 3-hydroxy-3'-4'-didehydro- β -caroten-4-one (HDCO). The monocyclic carotenoid torulene and HDCO, normally produced as minority carotenoids, were the main carotenoids produced in these strains.

L8 ANSWER 16 OF 24 EMBASE COPYRIGHT (c) 2005 Elsevier B.V. All rights reserved on STN

ACCESSION NUMBER: 2003499152 EMBASE
TITLE: Metabolic engineering of the astaxanthin-biosynthetic pathway of *Xanthophyllomyces dendrorhous*.
AUTHOR: Visser H.; Van Ooyen A.J.J.; Verdoes J.C.
CORPORATE SOURCE: H. Visser, Section of Fungal Genomics, Wageningen University, Dreijenlaan 2, 6703 HA Wageningen, Netherlands. hans.visser@wur.nl
SOURCE: FEMS Yeast Research, (2003) Vol. 4, No. 3, pp. 221-231.
Refs: 66
ISSN: 1567-1356 CODEN: FYREAG
COUNTRY: Netherlands
DOCUMENT TYPE: Journal; (Short Survey)
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 20040116
Last Updated on STN: 20040116

AB This review describes the different approaches that have been used to manipulate and improve carotenoid production in *Xanthophyllomyces dendrorhous*. The red yeast *X. dendrorhous* (formerly known as *Phaffia rhodozyma*) is one of the microbiological production systems for natural astaxanthin. Astaxanthin is applied in food and feed industry and can be used as a nutraceutical because of its strong antioxidant properties. However, the production levels of astaxanthin in wild-type isolates are rather low. To increase the astaxanthin content in *X. dendrorhous*, cultivation protocols have been optimized and astaxanthin-hyperproducing mutants have been

obtained by screening of classically mutagenized *X. dendrorhous* strains. The knowledge about the regulation of carotenogenesis in *X. dendrorhous* is still limited in comparison to that in other carotenogenic fungi. The *X. dendrorhous* carotenogenic genes have been cloned and a *X. dendrorhous* transformation system has been developed. These tools allowed the directed genetic modification of the **astaxanthin** pathway in *X. dendrorhous*. The *crtYB* gene, encoding the bifunctional enzyme phytoene synthase/lycopene cyclase, was inactivated by insertion of a vector by single and double cross-over events, indicating that it is possible to generate specific carotenoid-biosynthetic mutants. Additionally, overexpression of *crtYB* resulted in the accumulation of β -carotene and echinone, which indicates that the oxygenation reactions are rate-limiting in these recombinant strains. Furthermore, overexpression of the phytoene desaturase-encoding gene (*crtI*) showed an increase in monocyclic carotenoids such as torulene and HDCO (3-hydroxy-3',4'-didehydro- β , ψ -carotene-4-one) and a decrease in bicyclic carotenoids such as echinone, β -carotene and **astaxanthin**.
.COPYRGHT. 2003 Federation of European Microbiological Societies.
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L8 ANSWER 17 OF 24 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 2002-12964 BIOTECHDS

TITLE: Producing carotenoid compounds e.g. antheraxanthin and **astaxanthin**, by using microorganisms having a nucleic acid molecule encoding enzymes in the carotenoid biosynthetic pathway and which metabolize single carbon substrates; carotenoid production via bacterium, fungus host cell transformation using enzyme for the steroid production, flavor and electro-optic compound

AUTHOR: BRZOSTOWICZ P C; CHENG Q; DICOSIMO D J; KOFFAS M; MILLER E S; ODOM J M; PICATAGGIO S K; ROUVIERE P E

PATENT ASSIGNEE: DU PONT DE NEMOURS and CO E I

PATENT INFO: WO 2002018617 7 Mar 2002

APPLICATION INFO: WO 2000-US27420 1 Sep 2000

PRIORITY INFO: US 2000-229907 1 Sep 2000

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2002-351711 [38]

AB DERWENT ABSTRACT:

NOVELTY - Producing (M) a carotenoid compound, comprising providing a transformed C1 metabolizing host cell, comprising suitable levels of isopentenyl pyrophosphate and a nucleic acid molecule encoding an enzyme in the carotenoid biosynthetic pathway, under the control of regulatory sequences, and contacting the host cell with C1 carbon substrate to produce a carotenoid compound, is new.

DETAILED DESCRIPTION - An INDEPENDENT CLAIM is also included for over-production (M1) of a carotenoid in a transformed C1 metabolizing host cell, comprising suitable levels of isopentenyl pyrophosphate and a nucleic acid molecule encoding an enzyme in the carotenoid biosynthetic pathway, under the control of regulatory sequences, and contacting the host cell with C1 carbon substrate to produce a carotenoid compound.

BIOTECHNOLOGY - Preferred Method: In (M) the C1 carbon substrate is methane, methanol, formaldehyde, formic acid, methylated amines, methylated thiols or carbon dioxide. The C1 metabolizing host cell is a methylotroph or methanotroph chosen from *Methylomonas*, *Methylobacter*, *Methylococcus*, *Methylosinus*, *Methylocystis*, *Methylomicrobium*, *Methanomonas*, *Methylophilus*, *Methylobacillus*, *Methylobacterium*, *Hyphomicrobium*, *Xanthobacter*, *Bacillus*, *Paracoccus*, *Nocardia*, *Arthrobacter*, *Rhodopseudomonas*, *Pseudomonas*, *Candida*, *Hansenula*, *Pichia*, *Torulopsis* and *Rhodotorula*. The methanotroph is a high growth methanotropic strain which comprises a functional Embden-Meyerhof carbon pathway, comprising a gene encoding a pyrophosphate dependent phosphofructokinase (PFK) enzyme. The gene encoding PFK is an isolated nucleic acid molecule encoding a sequence (S1) of 437 amino acids defined in the specification, its hybridizable sequence, a nucleic acid molecule comprising a nucleotide sequence encoding a polypeptide of 437 amino acids that has 63% identity based on the Smith-Waterman method of alignment when compared to a polypeptide having (S1), or complement of the above nucleic acids. The high growth methanotrophic bacterial strain

optionally contains a gene encoding a fructose biphosphate aldolase enzyme, and functional Entner-Doudoroff carbon pathway, or a gene encoding a keto-deoxy phosphogluconate aldolase. The isolated nucleic acid molecule encodes a carotenoid biosynthetic enzyme such as geranylgeranyl pyrophosphate (GGPP) synthase, phytoene synthase, phytoene desaturase, lycopene cyclase, beta-carotene hydroxylase, zeaxanthin glucosyl transferase, beta-carotene ketolase, beta-carotene C-4 oxygenase, beta-carotene desaturase, spheroidene monooxygenase, carotene hydratase, carotenoid 3,4-desaturase, 1-OH-carotenoid methylase, farnesyl diphosphate synthetase, and diapophytoene dehydrogenase (whose sequences and Genbank Accession numbers are given in the specification). The nucleic acid molecule encoding GGPP synthase has a fully defined sequence of 303 amino acids and is chosen from Genbank Acc. such as AB000835, AB016043, AB019036, AB027705, AB027706, AB016044, AB034249, AB034250, AF020041, AF049658, AF049659, AF139916, AF279807, AF279808, AJ010302, AJ133724, AJ276129, D85029, L25813, L37405, U15778, U44876, X92893, X95596, X98795 and Y15112. Phytoene synthase comprises a sequence of 296 amino acids and is chosen from Genbank Acc AB001284, AB032797, AB034704, AB037975, AF009954, AF139916, AF152892, AF218415, AF220218, AJ010302, AJ133724, AJ278287, AJ304825, AJ308385, D58420, L23424, L25812, L37405, M38424, M87280, S71770, U32636, U62808, U87626, U91900, X52291, X60441, X63873, X68017, X69172 and X78814. Phytoene desaturase has a sequence of 492 amino acids and is chosen from Genbank Acc AB046992, AF039585, AF049356, AF139916, AF218415, AF251014, AF364515, D58420, D83514, L16237, L37405, M64704, M88683, S71770, U37285, U46919, U62808, X55289, X59948, X62574, X68058, X71023, X78271, X78434, X78815, X86783, Y14807, Y15007, Y15112, Y15114 and Z11165. Lycopene cyclase has a sequence of 382 amino acids defined in the specification and is chosen from Genbank Acc AF139916, AF152246, AF218415, AF272737, AJ13374, AJ250827, AJ276965, D58420, D83513, L40176, M87280, U50738, U50739, U62808, X74599, X81787, X86221, X95596 and X98796. beta-carotene hydroxylase has a sequence of 175 amino acids and is chosen from Genbank Acc D58420, D58422, D90087, M87280, U62808 and Y15112. Zeaxanthin glucosyl transferase has a sequence of 431 amino acids and is chosen from Genbank Acc D90087, M87280, and M90698. beta-carotene ketolase comprises a sequence of 532 amino acids and is chosen from Genbank Acc AF218415, D45881, D58420, D58422, X86782 and Y15112. In (M) and (M1) suitable levels of isopentenyl pyrophosphate are provided by the expression of heterologous upper pathway isoprenoid pathway genes such as D-1-deoxyxylulose-5-phosphate synthase (Dxs), D-1-deoxyxylulose-5-phosphate reductoisomerase (Dxr), 2C-methyl-d-erythritol cytidyltransferase (IspD), 4-diphosphocytidyl-2-C-methylerythritol kinase (IspE), 2C-methyl-d-erythritol 2,4-cyclodiphosphate synthase (IspF), CTP synthase (PyrG), IytB and GcpE. The gene encoding Dxs, Dxr, IspD, IspE, IspF, PyrG or IytB encodes a polypeptide of a sequence of 620, 394, 231, 285, 157, 544 or 318 amino acids, respectively defined in the specification.

USE - The method is useful for producing carotenoid compounds such as antheraxanthin, adonixanthin, **astaxanthin**, canthaxanthin, capsorubrin, beta-cryptoxanthin, alpha-carotene, beta-carotene, epsilon-carotene, echinenone, gamma-carotene, zeta-carotene, alpha-cryptoxanthin, diatoxanthin, 7,8-didehydroastaxanthin, fucoxanthin, fucoxanthinol, isorenieratene, lactucaxanthin, lutein, lycopene, neoxanthin, neurosporene, hydroxyneurosporene, peridinin, phytoene, rhodopin, rhodopin glucoside, siphonaxanthin, spheroidene, spheroidenone, spirilloxanthin, uriolide, uriolide acetate, violaxanthin, zeaxanthin-beta-digluconide and zeaxanthin (claimed). The carotenoids have potent anti-oxidant properties useful in diet, and aquaculture elements. The carotenoids are also useful as intermediates in the synthesis of steroids, flavors and fragrances and compounds for potential electro-optic applications.

EXAMPLE - The crt gene cluster comprising the crtEXYIBZ genes (crtE encoding geranylgeranyl pyrophosphate, crtX encoding zeaxanthin glycosyltransferase, crtY encoding lycopene cyclase, crtL encoding phytoene desaturase, crtB encoding phytoene synthase, crtZ encoding beta-carotene hydroxylase) from *Pantoea stewartii* was introduced into *Methylomonas* 16a to enable the synthesis of desirable 40-carbon carotenoids. Primers were designed using the sequence from *Erwinia uredovora* to amplify a fragment by polymerase chain reaction containing

the crt genes. These sequences included (I) and (II) and are denoted 5'-3'. For introduction into Methylobionas 16a, the crt gene cluster from pCR4-crt was first subcloned into the unique EcoRI site within the chloramphenicol-resistance gene of the broad host range vector, pBHR1. pBHR1 was linearized by digestion with EcoRI. pCR4-crt was digested with EcoRI and the 6.3 kb EcoRI fragment containing the crt gene cluster (crtEXYIB) was purified. This DNA fragment was ligated to EcoRI-digested pBHR1 and the ligated DNA was used to transform Escherichia coli DH5alpha by electroporation. Transformants were selected on Luria Bertani (LB) medium containing 50 microg/ml kanamycin. Several isolates were found to be sensitive to chloramphenicol and demonstrated a yellow colony phenotype after overnight incubation at 37degreesC. Analysis of the plasmid DNA from these transformants confirmed the presence of the crt gene cluster cloned in the same orientation as the pBHR1 chloramphenicol-resistance gene and this plasmid was designated pCrt1. Plasmid pCrt1 was transferred into Methylobionas 16a by tri-parental conjugate mating. E.coli helper strain containing pRK2013 and the E.coli DH5 alpha donor strain containing pCrt1 were grown overnight in LB medium containing kanamycin. The Methylobionas 16a recipient was grown for 48 hours in Nitrate liquid BTZ-3 medium containing 25% volume/volume methane. The donor, helper, and recipient cell pastes were combined on the surface of BTZ-3 agar plates containing 0.5% weight/volume yeast extract in ratios of 1:1:2 respectively. Plates were maintained at 30degreesC in 25% methane for 16-72 hours to allow conjugation to occur, after which the cell pastes were collected and resuspended in BTZ-3. For analysis of carotenoid composition, transconjugants were cultured in 25 ml BTZ-3 containing kanamycin (50 microg/ml) and incubated at 30degreesC in 25% methane as the sole carbon source. After thawing, the pellets were extracted and carotenoid content was analyzed by high performance liquid chromatography (HPLC). HPLC analysis of extracts from Methylobionas 16a containing pCrt1 confirmed the synthesis of beta-carotene. A single peak was present at 15.867 minutes, indicative of beta-carotene in the cultures. ATGACGGTCTGCGCAAAAAACACG (I) GAGAAATTATGTTGTGGATTGGAATGC (II) (156 pages)

L8 ANSWER 18 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2002:946472 HCAPLUS

DOCUMENT NUMBER: 138:23762

TITLE: Genes and enzymes of carotenoid biosynthesis of Paracoccus and their use in increasing yields in carotenoid fermentation

INVENTOR(S): Berry, Alan; Bretzel, Werner; Huembelin, Markus; Lopez-Ulibarri, Rual; Mayer, Anne Francoise; Yeliseev, Alexei

PATENT ASSIGNEE(S): Roche Vitamins AG, Switz.

SOURCE: PCT Int. Appl., 307 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002099095	A2	20021212	WO 2002-EP6171	20020605
WO 2002099095	A3	20031218		
W:	AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW			
RW:	GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG			
CA 2449885	AA	20021212	CA 2002-2449885	20020605
US 2003148416	A1	20030807	US 2002-166225	20020605
EP 1392824	A2	20040303	EP 2002-745362	20020605

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
IE, SI, LT, LV, FI, RO, MK, CY, AL, TR

JP 2004527265 T2 20040909 JP 2003-502205 20020605
PRIORITY APPLN. INFO.: US 2001-296299P P 20010606
WO 2002-EP6171 W 20020605

AB Genes of *Paracoccus* encoding enzymes of the mevalonate pathway of carotenoid biosynthesis, i.e. for HMG CoA reductase, isopentenyl diphosphate isomerase, hydroxymethylglutaryl-CoA synthase, mevalonate kinase, phosphomevalonate kinase, or diphosphomevalonate decarboxylase; are cloned and characterized for use in the construction of strains for the fermentation of isoprenoid compds. such as carotenoids like phytoene, lycopene, β -carotene, zeaxanthin, canthaxanthin, astaxanthin, adonixanthin, cryptoxanthin, echinenone and adonirubin. Expression vectors, cultured cells, and methods of making isoprenoid compds. are also provided. The strains of *Paracoccus* of interest have recently been reclassified from *Flavobacterium* on the basis of mol. and metabolic markers. Genes of the mevalonate biosynthetic cluster were cloned by PCR. Primers derived from highly conserved regions of known genes were used to obtain a fragment of the cluster that was used to identify flanking regions.

L8 ANSWER 19 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2003:811116 HCAPLUS

DOCUMENT NUMBER: 140:158546

TITLE: Recombinant expression of *Paracoccus marcusii* carotenoid biosynthesis-related gene in transgenic plant and *Escherichia coli*

INVENTOR(S): Yao, Quanhong; Peng, Rihe; Xiang, Aisheng

PATENT ASSIGNEE(S): Yongye Agricultural Science Biological Engineering Co., Ltd., Shanghai, Peop. Rep. China; Biological Technology Research Center, Shanghai Academy of Agricultural Sciences

SOURCE: Faming Zhuanli Shenqing Gongkai Shuomingshu, 36 pp.
CODEN: CNXXEV

DOCUMENT TYPE: Patent

LANGUAGE: Chinese

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
CN 1380415	A	20021120	CN 2001-105878	20010406
PRIORITY APPLN. INFO.:			CN 2001-105878	20010406

AB The protein and DNA sequences of three kinds of chimeric genes of crtWcrty, crtIcrtZ, and crtBcrtE of *Paracoccus marcusii*, which may induce the biosynthesis of astaxanthin, are presented. The gene crtE encodes geranylgeranyl pyrophosphate synthetase; gene crtB encodes phytoene synthetase; gene crtI encodes phytoene desaturase; gene crtZ encodes beta-carotene hydroxylase; gene crtY encodes lycopene beta-cyclase; and gene crtW encodes α -carotene ketolase. The invention also provides methods, vectors, restriction enzymes for construction of the three chimeric genes. The expression system may be used to improve plant quality and produce astaxanthin in plant.

L8 ANSWER 20 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1999:722785 HCAPLUS

DOCUMENT NUMBER: 131:334139

TITLE: DNA sequences encoding enzymes involved in production of isoprenoids in *Phaffia rhodozyma*

INVENTOR(S): Hoshino, Tatsuo; Ojima, Kazuyuki; Setoguchi, Yutaka

PATENT ASSIGNEE(S): F. Hoffmann-La Roche A.-G., Switz.

SOURCE: Eur. Pat. Appl., 58 pp.

CODEN: EPXXDW

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
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EP 955363	A2	19991110	EP 1999-107413	19990426
EP 955363	A3	20040128		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				
CA 2270711	AA	19991106	CA 1999-2270711	19990504
NO 9902165	A	19991108	NO 1999-2165	19990504
MX 9904154	A	20000731	MX 1999-4154	19990504
AU 9926962	A1	19991118	AU 1999-26962	19990505
AU 762939	B2	20030710		
BR 9901403	A	20001017	BR 1999-1403	19990505
CN 1234445	A	19991110	CN 1999-106367	19990506
JP 2000050884	A2	20000222	JP 1999-126015	19990506
US 6284506	B1	20010904	US 1999-306595	19990506
US 2003054523	A1	20030320	US 2001-925388	20010809
US 6586202	B2	20030701		
US 2003190734	A1	20031009	US 2003-431846	20030508
US 6872556	B2	20050329		

PRIORITY APPLN. INFO.:

EP 1998-108210	A	19980506
US 1999-306595	A3	19990506
US 2001-925388	A3	20010809

AB The present invention is directed to an isolated DNA sequence coding for an enzyme involved in the mevalonate pathway or the pathway from isopentenyl pyrophosphate to farnesyl pyrophosphate, vector of plasmids comprising such DNA, hosts transformed by either such DNAs or vectors or plasmids, and a process for the production of isoprenoids and carotenoids by using such transformed cell lines. Thus, the genes encoding 3-hydroxy-3-methylglutaryl-CoA synthetase (hmc), 3-hydroxy-3-methylglutaryl-CoA reductase (hmg), mevalonate kinase (mvk), mevalonate pyrophosphate decarboxylase (mpd), and farnesyl pyrophosphate synthetase (fps) are isolated from *Phaffia rhodozyma* ATCC 96594 and their sequences provided. These enzymes can be used for the synthesis of isoprenoids or carotenoids, such as **astaxanthin**.

L8 ANSWER 21 OF 24 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN DUPLICATE 5

ACCESSION NUMBER: 1999:232718 BIOSIS
DOCUMENT NUMBER: PREV199900232718
TITLE: The effect of the herbicide glufosinate (BASTA) on **astaxanthin** accumulation in the green alga *Haematococcus pluvialis*.
AUTHOR(S): Aflalo, Claude; Bing, Wang; Zarka, Aliza; Boussiba, Sammy [Reprint author]
CORPORATE SOURCE: Microalgal Biotechnology Laboratory, Jacob Blaustein Institute for Desert Research, Ben Gurion University of the Negev, Sede- Boker Campus, Negev, 84990, Israel
SOURCE: Zeitschrift fuer Naturforschung Section C Journal of Biosciences, (Jan.-Feb., 1999) Vol. 54, No. 1-2, pp. 49-54. print.
ISSN: 0939-5075.
DOCUMENT TYPE: Article
LANGUAGE: English
ENTRY DATE: Entered STN: 17 Jun 1999
Last Updated on STN: 17 Jun 1999

AB The addition of 2.5 mM glufosinate ammonium (BASTA), a well known plant killer, to *Haematococcus pluvialis* culture efficiently inhibits cell growth, blocks the activity of glutamine synthetase (GS) and induces **astaxanthin** accumulation. Conversely, methionine-S-sulfoximine (MSX), a well known GS inhibitor, had no effect on neither these parameters. When GS activity was tested in vitro, MSX inhibited the activity at high concentrations (mM), while glufosinate was effective in the μ M range. We have found that in the presence of glufosinate, ammonia is excreted from the cells. Therefore, we suggest that this process enables *Haematococcus* cells to escape the potentially harmful effect of glufosinate. As a consequence of the inability to assimilate nitrogen, **astaxanthin** is accumulated. This situation resembles the response of *Haematococcus* cells to nitrogen starvation.

L8 ANSWER 22 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1998:126371 HCAPLUS
 DOCUMENT NUMBER: 128:203178
 TITLE: Using enzymes of carotenoid biosynthesis to alter the carotenoid content and fatty acid profile of seeds
 INVENTOR(S): Shewmaker, Christine K.
 PATENT ASSIGNEE(S): Calgene, Inc.; USA; Shewmaker, Christine K.
 SOURCE: PCT Int. Appl., 70 pp.
 CODEN: PIXXD2
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 2
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9806862	A1	19980219	WO 1997-US14035	19970808
W: AL, AU, BA, BB, BG, BR, CA, CN, CU, CZ, EE, GE, HU, IL, IS, JP, KP, KR, LC, LK, LR, LT, LV, MG, MK, MN, MX, NO, NZ, PL, RO, SG, SI, SK, SL, TR, TT, UA, US, UZ, VN, YU, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: GH, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
CA 2261577	AA	19980219	CA 1997-2261577	19970808
AU 9740584	A1	19980306	AU 1997-40584	19970808
EP 925366	A1	19990630	EP 1997-938203	19970808
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
CN 1227609	A	19990901	CN 1997-197150	19970808
BR 9713462	A	20000328	BR 1997-13462	19970808
JP 2001505409	T2	20010424	JP 1998-509911	19970808
ZA 9707469	A	19980219	ZA 1997-7469	19970820
US 2002092039	A1	20020711	US 2002-41472	20020110
PRIORITY APPLN. INFO.:			US 1996-24145P	P 19960809
			US 1997-908758	A1 19970808
			WO 1997-US14035	W 19970808

AB Methods of altering the carotenoid content and fatty acid profile of seeds by altering the levels of expression of genes for enzymes of carotenoid biosynthesis. Increasing the diversion of acetate to carotenoid biosynthesis increases the anti-oxidant content of the oil, lowers the level of oxidation-prone unsatd. fatty acids such as linoleate or linolenate, and increases the oleic acid content of the oil. Preferably, the enzyme is one of the earlier enzymes in the carotenoid pathway. The crtB gene of Erwinia uredovora, encoding phytoene synthase, was placed under control of a napin gene promoter using the signal sequence of the RuBisco small subunit gene and the construct introduced into Brassica napus by Agrobacterium-mediated transformation. T2 plants showed Mendelian segregation of an orange phenotype. Seed from these plants showed increased levels of carotenoids and tocopherols, with several carotenoids not detectable in control seeds being found in transgenic seed. The fatty acid composition of the seeds showed an increase in oleic acid content at the expense of linoleic and linolenic acid levels. Transgenic seeds showed slower germination than control seeds, but the germination rate was not affected.

REFERENCE COUNT: 16 THERE ARE 16 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L8 ANSWER 23 OF 24 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1998:394053 HCAPLUS
 DOCUMENT NUMBER: 129:94523
 TITLE: Recombinant preparation of carotenoids using enzymes from Flavobacterium or gram-negative bacteria strain E-396 for feed or food industries
 INVENTOR(S): Pasamontes, Luis; Tosigonkov, Juri
 PATENT ASSIGNEE(S): F. Hoffmann-La Roche A.-G., Switz.
 SOURCE: Jpn. Kokai Tokkyo Koho, 80 pp.
 CODEN: JKXXAF
 DOCUMENT TYPE: Patent
 LANGUAGE: Japanese

FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 10155497	A2	19980616	JP 1997-348653	19971202
EP 872554	A2	19981021	EP 1997-120324	19971120
EP 872554	A3	20000607		
EP 872554	B1	20030611		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				
AT 242812	E	20030615	AT 1997-120324	19971120
ES 2200110	T3	20040301	ES 1997-120324	19971120
BR 9705676	A	19990525	BR 1997-5676	19971201
US 6291204	B1	20010918	US 1997-980832	19971201
CN 1184159	A	19980610	CN 1997-122604	19971202
IN 190903	A	20030830	IN 2000-MA109	20000210
US 2003022273	A1	20030130	US 2001-920923	20010802
US 6677134	B2	20040113		
US 2004058410	A1	20040325	US 2003-695980	20031029
PRIORITY APPLN. INFO.:			EP 1996-810839	A 19961202
			US 1997-980832	A3 19971201
			IN 1998-MA2511	A 19981106
			US 2001-920923	A3 20010802

AB Disclosed is a method for industrial-scale production of carotenoids by expression of the Flavobacterium strain R1534- or gram-neg. bacteria strain E-396-derived genes that are associated with the carotenoids-biosynthesis in a transgenic host such as Escherichia coli or Bacillus subtilis. The genes involved are crtE (for geranylgeranyl pyrophosphate synthetase), crtB (phytoene synthetase), crtI (phytoene desaturase), crtY (lycopene cyclase), all from Flavobacterium strain R1534, and crtZE396 (β -carotene oxygenase) from gram-neg. bacteria strain E-396. Gene crtW encoding β -carotene β 4-oxygenase of Alcaligenes strain PC-1 may also be used to improve the carotenoids production. Methods for fermentation production of cantaxanthin, astaxanthin, adonixanthin, and zeaxanthin are claimed. Methods using genes crtEE396, crtBE396, crtIE396, crtYE396, crtZE396, and crtWE396, all from gram-neg. bacteria strain E-396, also claimed. Use of carotenoids as food or feed additives is also claimed.

L8 ANSWER 24 OF 24 EMBASE COPYRIGHT (c) 2005 Elsevier B.V. All rights reserved on STN DUPLICATE 6

ACCESSION NUMBER: 94161961 EMBASE
DOCUMENT NUMBER: 1994161961
TITLE: β -carotene producing mutants of Phaffia rhodozyma.
AUTHOR: Girard P.; Falconnier B.; Bricout J.; Vladescu B.
CORPORATE SOURCE: Laboratoire de Microbiologie, Centre de Recherche Pernod-Ricard, 120 Avenue du Marechal Foch, 94015 Creteil Cedex, France
SOURCE: Applied Microbiology and Biotechnology, (1994) Vol. 41, No. 2, pp. 183-191.
ISSN: 0175-7598 CODEN: AMBIDG
COUNTRY: Germany
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 940629
Last Updated on STN: 940629

AB Like other carotenoid-producing organisms, Phaffia rhodozyma, a red astaxanthin-producing yeast, is supposed to synthesize carotenoids by the following steps: formation of phytoene from geranylgeranyl pyrophosphate, dehydrogenation of phytoene to lycopene, cyclization of lycopene to β -carotene and oxidation of the latter to astaxanthin. Mutagenic treatments generated in P. rhodozyma a wide diversity of colour variants ranging from white to dark red. The identification of the corresponding carotenoid compounds revealed the occurrence of β -carotene-accumulating strains, phytoene-accumulating strains, and strains lacking any carotenoid compound. These classes of

strains are likely to result from alterations in, respectively, the oxidation of β -carotene, phytoene dehydrogenation and the phytoene synthetase step. Except for the cyclization of lycopene to β -carotene, all the steps of carotenogenesis in *P. rhodozyma* are represented by the above mutants. Furthermore, astaxanthin-overproducing mutants were also selected; they are likely to be affected in some upstream step, and certainly before β -carotene, as after an additional mutagenesis they generated oxidaseless strains that, in this case, overproduce β -carotene. The latter strains appear very promising for biotechnological production of natural β -carotene.

=> d his

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

```
L1      4 S ASTAXANTHIN (W) SYNTHASE?
L2      0 S ASTAXANTHIN (W) SYNTHETASE?
L3     6416 S ASTAXANTHIN
L4     24046 S "AST"
L5    194127 S SYNTHETASE?
L6      0 S L3 (W) L5
L7     39 S L3 AND L5
L8     24 DUP REM L7 (15 DUPLICATES REMOVED)
```

=> s reaction (a) center

```
L9      41050 REACTION (A) CENTER
```

=> s l8 and l9

```
L10     0 L8 AND L9
```

=> s l5 and l9

```
L11     153 L5 AND L9
```

=> s reductase?

```
L12     371127 REDUCTASE?
```

=> s l11 and l12

```
L13     21 L11 AND L12
```

=> dupo rem l13

DUPO IS NOT A RECOGNIZED COMMAND

The previous command name entered was not recognized by the system.
For a list of commands available to you in the current file, enter
"HELP COMMANDS" at an arrow prompt (=>).

=> dup rem l13

PROCESSING COMPLETED FOR L13

```
L14     19 DUP REM L13 (2 DUPLICATES REMOVED)
```

=> d 1-19 ibib ab

L14 ANSWER 1 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2005:83313 HCAPLUS

DOCUMENT NUMBER: 142:294593

TITLE: Roseobacter-like bacteria in red and Mediterranean Sea
aerobic anoxygenic photosynthetic populations

AUTHOR(S): Oz, Aia; Sabehi, Gazalah; Koblizek, Michal; Massana,
Ramon; Beja, Oded

CORPORATE SOURCE: Department of Biology, Technion-Israel Institute of
Technology, Haifa, Israel

SOURCE: Applied and Environmental Microbiology (2005), 71(1),
344-353

CODEN: AEMIDF; ISSN: 0099-2240

PUBLISHER: American Society for Microbiology

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Bacteriochlorophyll a-containing aerobic anoxygenic phototrophs (AAnP) have been proposed to account for up to 11% of the total surface water microbial community and to potentially have great ecol. importance in the world's oceans. Recently, environmental and genomic data based on anal. of the *pufM* gene identified the existence of α -proteobacteria as well as possible γ -like proteobacteria among AAnP in the Pacific Ocean. Here we report on analyses of environmental samples from the Red and Mediterranean Seas by using *pufM* as well as the *bchX* and *bchL* genes as mol. markers. The majority of photosynthesis genes retrieved from these seas were related to Roseobacter-like AAnP sequences. Furthermore, the sequence of a novel photosynthetic operon organization from an uncultured Roseobacter-like bacterial artificial chromosome retrieved from the Red Sea is described. The data show the presence of Roseobacter-like bacteria in Red and Mediterranean Sea AAnP populations in the seasons analyzed.

REFERENCE COUNT: 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 2 OF 19 MEDLINE on STN
ACCESSION NUMBER: 2004496572 MEDLINE
DOCUMENT NUMBER: PubMed ID: 15466032
TITLE: The glutathione-glutaredoxin system in *Rhodobacter capsulatus*: part of a complex regulatory network controlling defense against oxidative stress.
AUTHOR: Li Kuanyu; Hein Silke; Zou Wenxin; Klug Gabriele
CORPORATE SOURCE: Institut fur Mikrobiologie und Molekularbiologie, Heinrich-Buff-Ring 26-32, 35392 Giessen, Germany.
SOURCE: Journal of bacteriology, (2004 Oct) 186 (20) 6800-8.
Journal code: 2985120R. ISSN: 0021-9193.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200411
ENTRY DATE: Entered STN: 20041007
Last Updated on STN: 20041103
Entered Medline: 20041102

AB Mutants with defects in components of the glutathione-glutaredoxin (GSH/Grx) system of *Rhodobacter capsulatus* were constructed to study its role in defense against oxidative stress and the redox-dependent formation of photosynthetic complexes. The lack of the glutaredoxin 3 gene (*grxC*) or the glutathione synthetase B gene (*gshB*) resulted in lower growth rates under aerobic conditions and higher sensitivity to oxidative stress, confirming the role of the GSH/Grx system in oxidative stress defense. Both mutants are highly sensitive to disulfide stress, indicating a major contribution of the GSH/Grx system to the thiol-disulfide redox buffer in the cytoplasm. Like mutations in the thioredoxin system, mutations in the GSH/Grx system affected the formation of photosynthetic complexes, which is redox dependent in *R. capsulatus*. Expression of the genes *grxC*, *gshB*, *grxA* for glutaredoxin 1, and *gorA* for glutathione reductase, all encoding components of the GSH/Grx system, was not induced by oxidative stress. Other genes, for which a role in oxidative stress was established in *Escherichia coli*, *acnA*, *fpr*, *fur*, and *katG*, were strongly induced by oxidative stress in *R. capsulatus*. Mutations in the *grxC*, and/or *gshB*, and/or *trxC* (thioredoxin 2) genes affected expression of these genes, indicating an interplay of the different defense systems against oxidative stress. The OxyR and the SoxRS regulons control the expression of many genes involved in oxidative stress defense in *E. coli* in response to H₂O₂ and superoxide, respectively. Our data and the available genome sequence of *R. capsulatus* suggest that a SoxRS system is lacking but an alternative superoxide specific regulator exists in *R. capsulatus*. While the expression of *gorA* and *grxA* is regulated by H₂O₂ in *E. coli* this is not the case in *R. capsulatus*, indicating that the OxyR regulons of these two species are significantly different.

L14 ANSWER 3 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 2004:878802 HCAPLUS
DOCUMENT NUMBER: 142:170748
TITLE: Comparative analysis of the complete plastid genome

sequence of the red alga *Gracilaria tenuistipitata* var. *liui* provides insights into the evolution of rhodoplasts and their relationship to other plastids
AUTHOR(S): Hagopian, Jonathan C.; Reis, Marcelo; Kitajima, Joao P.; Bhattacharya, Debashish; de Oliveira, Mariana C.
CORPORATE SOURCE: Departamento de Botanica, Instituto de Biociencias, Universidade de Sao Paulo, Sao Paulo, 05508-900, Brazil
SOURCE: Journal of Molecular Evolution (2004), 59(4), 464-477
CODEN: JMEVAU; ISSN: 0022-2844
PUBLISHER: Springer New York, LLC
DOCUMENT TYPE: Journal
LANGUAGE: English

AB We sequenced to completion the circular plastid genome of the red alga *Gracilaria tenuistipitata* var. *liui*. This is the first plastid genome sequence from the subclass Florideophycidae (Rhodophyta). The genome is composed of 183,883 bp and contains 238 predicted genes, including a single copy of the rRNA operon. Comparisons with the plastid genome of *Porphyra pupurea* reveal strong conservation of gene content and order, but we found major genomic rearrangements and the presence of coding regions that are specific to *Gracilaria*. Phylogenetic anal. of a data set of 41 concatenated proteins from 23 plastid and two cyanobacterial genomes support red algal plastid monophyly and a specific evolutionary relationship between the Florideophycidae and the Bangiales. *Gracilaria* maintains a surprisingly ancient gene content in its plastid genome and, together with other Rhodophyta, contains the most complete repertoire of plastid genes known in photosynthetic eukaryotes.

REFERENCE COUNT: 24 THERE ARE 24 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 4 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2004:746079 HCAPLUS
DOCUMENT NUMBER: 141:361356
TITLE: High levels of RNA editing in a vascular plant chloroplast genome: analysis of transcripts from the fern *Adiantum capillus-veneris*
AUTHOR(S): Wolf, Paul G.; Rowe, Carol A.; Hasebe, Mitsuyasu
CORPORATE SOURCE: Department of Biology, Utah State University, Logan, UT, 84322, USA
SOURCE: Gene (2004), 339, 89-97
CODEN: GENED6; ISSN: 0378-1119
PUBLISHER: Elsevier B.V.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB We sequenced transcripts from all putative genes for proteins, rRNAs, and a selection of gene-encoding tRNAs in the chloroplast genome of the fern *Adiantum capillus-veneris*. We detected 350 RNA editing sites when the cDNA sequence was compared to that of the genomic DNA. Of these sites, 10% were U-to-C edits and 90% were C-to-U edits. RNA editing created 19 new start codons, three new stop codons, and "repaired" 26 internal stop codons. Of the 332 editing sites that altered a codon, 26% were in the first codon position, 68% in the second, and 6% in the third. We also detected 21 silent edits, as well as 19 edits that were in untranslated regions, including introns and the anticodon of tRNA^{Leu}. The latter edit provided a tRNA that is not otherwise encoded in this genome and accounts for a heavily used leucine codon. The level of RNA editing in this fern is more than ten times that of any other vascular plant examined across an entire chloroplast genome. A previous study found even higher levels of editing in a hornwort (942 sites). This suggests that the relatively low levels of editing in seed plants (less than 0.05%) may not be typical for land plants, and that RNA editing may play a major role in chloroplast genome processing. Addnl., we found that 53 editing sites in *A. capillus-veneris* are homologous to editing sites in the hornwort, and some other land plants. This implies that a major component of RNA editing sites have been conserved for hundreds of millions of years.

REFERENCE COUNT: 29 THERE ARE 29 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 5 OF 19 MEDLINE on STN

ACCESSION NUMBER: 2003129361 MEDLINE
DOCUMENT NUMBER: PubMed ID: 12643044
TITLE: Mechanisms of metalloenzymes studied by quantum chemical methods.
AUTHOR: Siegbahn Per E M
CORPORATE SOURCE: Department of Physics, Stockholm Centre for Physics, Astronomy and Biotechnology (SCFAB), Stockholm University, S-106 91 Stockholm, Sweden.
SOURCE: Quarterly reviews of biophysics, (2003 Feb) 36 (1) 91-145.
Ref: 125
Journal code: 0144032. ISSN: 0033-5835.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200309
ENTRY DATE: Entered STN: 20030320
Last Updated on STN: 20030928
Entered Medline: 20030926

AB The study of metalloenzymes using quantum chemical methods of high accuracy is a relatively new field. During the past five years a quite good understanding has been reached concerning the methods and models to be used for these systems. For systems containing transition metals hybrid density functional methods have proven both accurate and computationally efficient. A background on these methods and the accuracy achieved in benchmark tests are given first in this review. The rest of the review describes examples of studies on different metalloenzymes. Most of these examples describe mechanisms where dioxygen is either formed, as in photosystem II, or cleaved as in many other enzymes like cytochrome c oxidase, ribonucleotide reductase, methane mono-oxygenase and tyrosinase. In the descriptions below high emphasis is put on the actual determination of the transition states, which are the key points determining the mechanisms.

L14 ANSWER 6 OF 19 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 2002:203471 SCISEARCH
THE GENUINE ARTICLE: 526LK
TITLE: Rhodospirillum rubrum possesses a variant of the bchP gene, encoding geranylgeranyl-bacteriopheophytin reductase
AUTHOR: Addlesee H A; Hunter C N (Reprint)
CORPORATE SOURCE: Univ Sheffield, Krebs Inst, Dept Mol Biol & Biotechnol, Firth Court, Western Bank, Sheffield S10 2TN, S Yorkshire, England (Reprint); Univ Sheffield, Krebs Inst, Dept Mol Biol & Biotechnol, Sheffield S10 2TN, S Yorkshire, England; Univ Sheffield, Robert Hill Inst Photosynth, Dept Mol Biol & Biotechnol, Sheffield S10 2TN, S Yorkshire, England
COUNTRY OF AUTHOR: England
SOURCE: JOURNAL OF BACTERIOLOGY, (MAR 2002) Vol. 184, No. 6, pp. 1578-1586.
ISSN: 0021-9193.
PUBLISHER: AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC 20036-2904 USA.
DOCUMENT TYPE: Article; Journal
LANGUAGE: English
REFERENCE COUNT: 40
ENTRY DATE: Entered STN: 15 Mar 2002
Last Updated on STN: 15 Mar 2002

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The bchP gene product of Rhodobacter sphaeroides is responsible for the reduction of the isoprenoid moiety of bacteriochlorophyll (Bchl) from geranylgeraniol (GG) to phytol; here, we show that this enzyme also catalyzes the reduction of the isoprenoid moiety of bacteriopheophytin (Bphe). In contrast, we demonstrate that a newly identified homolog of this gene in Rhodospirillum rubrum encodes an enzyme, GG-Bphe reductase, capable of reducing the isoprenoid moiety of Bphe only.

We propose that *Rhodospirillum rubrum* is a naturally occurring bchP mutant and that an insertion mutation may have been the initial cause of a partial loss of function. Normal BchP function can be restored to *Rhodospirillum rubrum*, creating a new transconjugant strain possessing Bchl esterified with phytol. We speculate on the requirement of *Rhodospirillum rubrum* for phytylated Bphe and on a potential link between the absence of LH2 and of phytylated Bchl from the wild-type bacterium. The identification of a second role for the fully functional BchP in catalyzing the synthesis of phytylated Bphe strongly suggests that homologs of this enzyme may be similarly responsible for the synthesis of phytylated pheophytin in organisms possessing photosystem 2. In addition to bchP, other members of a photosynthesis gene cluster were identified in *Rhodospirillum rubrum*, including a bchG gene, demonstrated to encode a functional Bchl synthetase by complementation of a *Rhodobacter sphaeroides* mutant.

L14 ANSWER 7 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2001:280124 HCAPLUS
DOCUMENT NUMBER: 136:1395
TITLE: Horizontal Transfer of the Photosynthesis Gene Cluster and Operon Rearrangement in Purple Bacteria
AUTHOR(S): Igarashi, Naoki; Harada, Jiro; Nagashima, Sakiko; Matsura, Katsumi; Shimada, Keizo; Nagashima, Kenji V. P.
CORPORATE SOURCE: Department of Biology, Minamiohsawa 1-1, Tokyo Metropolitan University, Hachioji, Tokyo, 192-0397, Japan
SOURCE: Journal of Molecular Evolution (2001), 52(4), 333-341
CODEN: JMEVAU; ISSN: 0022-2844
PUBLISHER: Springer-Verlag New York Inc.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB A 37-kb photosynthesis gene cluster was sequenced in a photosynthetic bacterium belonging to the β subclass of purple bacteria (Proteobacteria), *Rubrivivax gelatinosus*. The cluster contained 12 bacteriochlorophyll biosynthesis genes (bch), 7 carotenoid biosynthesis genes (crt), structural genes for photosynthetic apparatuses (puf and puh), and some other related genes. The gene arrangement was markedly different from those of other purple photosynthetic bacteria, while two superoperonal structures, crtEF-bchCXYZ-puf and bchFNBHLM-lhaA-puhA, were conserved. Mol. phylogenetic analyses of these photosynthesis genes showed that the photosynthesis gene cluster of *R. gelatinosus* was originated from those of the species belonging to the α subclass of purple bacteria. It was concluded that a horizontal transfer of the photosynthesis gene cluster from an ancestral species belonging to the α subclass to that of the β subclass of purple bacteria had occurred and was followed by rearrangements of the operons in this cluster.

REFERENCE COUNT: 42 THERE ARE 42 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 8 OF 19 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 2001:759593 SCISEARCH
THE GENUINE ARTICLE: 474QH
TITLE: Regulation of catalytic activity of a multifunctional polyketide biosynthetic enzyme, 6-hydroxymellein synthase, by interaction between NADPH and phenylglyoxal-sensitive amino acid residue at the reaction center
AUTHOR: Kurosaki F (Reprint); Togashi K; Arisawa M
CORPORATE SOURCE: Toyama Med & Pharmaceut Univ, Fac Pharmaceut Sci, Toyama 9300194, Japan (Reprint)
COUNTRY OF AUTHOR: Japan
SOURCE: BIOCHIMICA ET BIOPHYSICA ACTA-PROTEIN STRUCTURE AND MOLECULAR ENZYMOLOGY, (10 SEP 2001) Vol. 1549, No. 1, pp. 51-60.
ISSN: 0167-4838.
PUBLISHER: ELSEVIER SCIENCE BV, PO BOX 211, 1000 AE AMSTERDAM,

NETHERLANDS.

DOCUMENT TYPE: Article; Journal
LANGUAGE: English
REFERENCE COUNT: 30
ENTRY DATE: Entered STN: 5 Oct 2001
Last Updated on STN: 5 Oct 2001

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Treatment of 6-hydroxymellein synthase, a multifunctional polyketide biosynthetic enzyme in carrot cells, with phenylglyoxal yielded a chemically modified protein in which approximately two moles of the reagent were covalently attached to each subunit of the enzyme. Only NADH- but not NADPH-associated form of native 6-hydroxymellein synthase was inhibited by cerulenin; however, the NADPH-synthase complex lost the insensitivity by the chemical modification of the enzyme protein with phenylglyoxal. Appreciable differences in K-m values observed between the NADPH- and NADH-associated enzymes were greatly reduced by the treatment with phenylglyoxal. Although the catalytic activity of the NADPH-associated synthase was enhanced by the addition of free CoA. the compound exhibited a significant inhibitory activity to the phenylglyoxal-modified enzyme, A marked deuterium isotope effect in the catalytic reaction of the native synthase-NADPH complex was appreciably decreased in the chemically modified enzyme. These results strongly suggest that an electrostatic interaction between the phosphate group attached to the 2' -position of adenosyl moiety of NADPH and the phenylglyoxal-sensitive amino acid residue, probably arginine, at the reaction center of 6-hydroxymellein synthase regulates several biochemical properties of this multifunctional enzyme. (C) 2001 Elsevier Science BN. All rights reserved.

L14 ANSWER 9 OF 19 MEDLINE on STN

ACCESSION NUMBER: 2001434305 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11181712
TITLE: Chloroplast precursor proteins compete to form early import intermediates in isolated pea chloroplasts.
AUTHOR: Row P E; Gray J C
CORPORATE SOURCE: Department of Plant Sciences, University of Cambridge, Downing Street, Cambridge CB2 3EA, UK.
SOURCE: Journal of experimental botany, (2001 Jan) 52 (354) 47-56.
Journal code: 9882906. ISSN: 0022-0957.
PUB. COUNTRY: England: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200108
ENTRY DATE: Entered STN: 20010806
Last Updated on STN: 20010806
Entered Medline: 20010802

AB In order to ascertain whether there is one site for the import of precursor proteins into chloroplasts or whether different precursor proteins are imported via different import machineries, chloroplasts were incubated with large quantities of the precursor of the 33 kDa subunit of the oxygen-evolving complex (pOE33) or the precursor of the light-harvesting chlorophyll a/b-binding protein (pLHCP) and tested for their ability to import a wide range of other chloroplast precursor proteins. Both pOE33 and pLHCP competed for import into chloroplasts with precursors of the stromally-targeted small subunit of Rubisco (pSSu), ferredoxin NADP(+) reductase (pFNR) and porphobilinogen deaminase; the thylakoid membrane proteins LHCP and the Rieske iron-sulphur protein (pRieske protein); ferrochelatase and the gamma subunit of the ATP synthase (which are both associated with the thylakoid membrane); the thylakoid lumenal protein plastocyanin and the phosphate translocator, an integral membrane protein of the inner envelope. The concentrations of pOE33 or pLHCP required to cause half-maximal inhibition of import ranged between 0.2 and 4.9 microm. These results indicate that all of these proteins are imported into the chloroplast by a common import machinery. Incubation of chloroplasts with pOE33 inhibited the formation of early import intermediates of pSSu, pFNR and pRieske protein.

L14 ANSWER 10 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2000:831437 HCAPLUS
DOCUMENT NUMBER: 134:338070
TITLE: The structure and gene repertoire of an ancient red algal plastid genome
AUTHOR(S): Glockner, Gernot; Rosenthal, Andre; Valentin, Klaus
CORPORATE SOURCE: IMB Jena, Dept. of Genome Analysis, Jena, 07745, Germany
SOURCE: Journal of Molecular Evolution (2000), 51(4); 382-390
CODEN: JMEVAU; ISSN: 0022-2844
PUBLISHER: Springer-Verlag New York Inc.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Photosynthetic eukaryotes can, according to features of their chloroplasts, be divided into two major groups: the red and the green lineage of plastid evolution. To extend the knowledge about the evolution of the red lineage we have sequenced and analyzed the chloroplast genome (cp-genome) of *Cyanidium caldarium* RK1, a unicellular red alga (AF022186). The anal. revealed that this genome shows several unusual structural features, such as a hypothetical hairpin structure in a gene-free region and absence of large repeat units. We provide evidence that this structural organization of the cp-genome of *C. caldarium* may be that of the most ancient cp-genome so far described. We also compared the cp-genome of *C. caldarium* to the other known cp-genomes of the red lineage. The cp-genome of *C. caldarium* cannot be readily aligned with that of *Porphyra purpurea*, a multicellular red alga, or *Guillardia theta* due to a displacement of a region of the cp-genome. The phylogenetic tree reveals that the secondary endosymbiosis, through which *G. theta* evolved, took place after the separation of the ancestors of *C. caldarium* and *P. purpurea*. We found several genes unique to the cp-genome of *C. caldarium*. Five of them seem to be involved in the building of bacterial cell envelopes and may be responsible for the thermotolerance of the chloroplast of this alga. Two addnl. genes may play a role in stabilizing the photosynthetic machinery against salt stress and detoxification of the chloroplast. Thus, these genes may be unique to the cp-genome of *C. caldarium* and may be required for the endurance of the extreme living conditions of this alga.

REFERENCE COUNT: 41 THERE ARE 41 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 11 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2000:116075 HCAPLUS
DOCUMENT NUMBER: 132:304134
TITLE: The complete sequence of a heterochromatic island from a higher eukaryote
AUTHOR(S): McCombie, W. Richard; De la Bastide, Melissa; Habermann, Kristina; Parnell, Laurence; Dedhia, Neilay; Gnoj, Lidia; Schutz, Kristin; Huang, Emily; Spiegel, Lori; Yordan, Cristy; Sehkun, Mundeep; Murray, Jennifer; Sheet, Paul; Cordes, Matt; Threideh, Jane; Stoneking, Tamberlyn; Kalicki, Joelle; Graves, Tina; Harmon, Gwen; Edwards, Jennifer; Latreille, Phil; Courtney, Laura; Cloud, James; Abbott, Amanda; Scott, Kelsi; Johnson, Doug; Minx, Pat; Bentley, Dan; Fulton, Bob; Miller, Nancy; Greco, Tracie; Kemp, Kim; Kramer, Jason; Fulton, Lucinda; Mardis, Elaine; Dante, Mike; Pepin, Kym; Hillier, LaDeana; Nelson, Joanne; Spieth, John; Simorowski, Joe; May, Bruce; Ma, Peter; Preston, Ray; Vill, Daniel; See, Lei Hoon; Shekher, Monica; Matero, Anthony; Shah, Ravi; Swaby, I'Kyori; O'Shaughnessy, Andrew; Rodriguez, Milka; Hoffman, Jane; Till, Sally; Granat, Susan; Shohdy, Nadim; Hasegawa, Amy; Hameed, Aliyah; Lodhi, Mohammad; Johnson, Arthur; Chen, Ellson; Marra, Marco; Wilson, Richard K.; Martienssen, Robert
CORPORATE SOURCE: Cold Spring Harbor Laboratory, Lita Annenberg Hazen Genome Center, Cold Spring Harbor, NY, 11724, USA
SOURCE: Cell (Cambridge, Massachusetts) (2000), 100(3), 377-386
CODEN: CELLB5; ISSN: 0092-8674

PUBLISHER: Cell Press
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Heterochromatin, constitutively condensed chromosomal material, is widespread among eukaryotes but incompletely characterized at the nucleotide level. We have sequenced and analyzed 2.1 megabases (Mb) of Arabidopsis thaliana chromosome 4 that includes 0.5-0.7 Mb of isolated heterochromatin that resembles the chromosomal knobs described by Barbara McClintock in maize. This isolated region has a low d. of expressed genes, low levels of recombination and a low incidence of gene trap insertion. Satellite repeats were absent, but tandem arrays of long repeats and many transposons were found. Methylation of these sequences was dependent on chromatin remodeling. Clustered repeats were associated with condensed chromosomal domains elsewhere. The complete sequence of a heterochromatic island provides an opportunity to study sequence determinants of chromosome condensation.

REFERENCE COUNT: 90 THERE ARE 90 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 12 OF 19 MEDLINE on STN

ACCESSION NUMBER: 2000129779 MEDLINE

DOCUMENT NUMBER: PubMed ID: 10664131

TITLE: Leaf-specific overexpression of plastidic glutamine synthetase stimulates the growth of transgenic tobacco seedlings.

AUTHOR: Migge A; Carrayol E; Hirel B; Becker T W

CORPORATE SOURCE: Lehrstuhl fur Genetik, Fakultat fur Biologie, Universitat Bielefeld, Postfach 10 01 31, D-33501 Bielefeld, Germany.

SOURCE: Planta, (2000 Jan) 210 (2) 252-60.
Journal code: 1250576. ISSN: 0032-0935.

PUB. COUNTRY: GERMANY: Germany, Federal Republic of

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 200006

ENTRY DATE: Entered STN: 20000613

Last Updated on STN: 20000613

Entered Medline: 20000601

AB The impact of increased plastidic glutamine synthetase (GS-2; EC 6.1.3.2) activity on foliar amino-acid levels and on biomass production was examined in transgenic tobacco. For that, tobacco was transformed via Agrobacterium tumefaciens with a binary vector containing a tobacco GS-2 cDNA downstream of the leaf-specific soybean ribulose-1,5-bisphosphate carboxylase/oxygenase small subunit gene promoter. Two transgenic tobacco lines with 15- to 18-fold higher foliar GS-2 transcript levels than the wild type were obtained. The GS-2 protein pools and the specific GS-2 activities were, however, only 2- to 2.3-fold higher in the leaves of the transgenic plants than in the leaves of the wild type. This discrepancy may reflect a post-transcriptional control of GS-2 protein accumulation. The increased GS-2 activity was correlated with a decrease in the leaf ammonium pool (3.7-fold) and an increase in the levels of some free amino acids, including glutamate (2.5-fold) and glutamine (2.3-fold). The accumulation of soluble protein per unit fresh weight, however, remained unchanged. This result indicates that a process downstream of the synthesis of the primary organic products of N-assimilation is limiting leaf protein accumulation. Nevertheless, the overexpression of GS-2 stimulated the growth rate of the transgenic tobacco seedlings which, consequently, were larger (20-30% on a fresh-weight basis) than wild-type seedlings grown under identical conditions. This result suggests that GS-2 is the rate-limiting enzyme during biomass production in tobacco seedlings. The requirement for glutamate as the ammonium acceptor in the reaction catalysed by GS-2 may imply that there is co-regulation of GS-2 and ferredoxin dependent glutamate synthase (Fd-GOGAT; EC 1.4.7.1) gene expression. Increased leaf GS-2 activity had, however, no influence on the foliar Fd-GOGAT protein abundance. This result suggests that in tobacco leaves, more Fd-GOGAT is present than required to meet the demands of primary ammonium assimilation and that there is no strong interdependence between GS-2 and Fd-GOGAT protein expression.

L14 ANSWER 13 OF 19 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 2001:90274 SCISEARCH

THE GENUINE ARTICLE: 392XY

TITLE: Role of reducing co-factors in catalytic reactions of 6-hydroxymellein synthase, a multifunctional polyketide biosynthetic enzyme in carrot cells

AUTHOR: Kurosaki F (Reprint); Togashi K; Arisawa M

CORPORATE SOURCE: Toyama Med & Pharmaceut Univ, Fac Pharmaceut Sci, Toyama 9300194, Japan (Reprint)

COUNTRY OF AUTHOR: Japan

SOURCE: PLANT SCIENCE, (7 DEC 2000) Vol. 160, No. 1, pp. 113-120. ISSN: 0168-9452.

PUBLISHER: ELSEVIER SCI IRELAND LTD, CUSTOMER RELATIONS MANAGER, BAY 15, SHANNON INDUSTRIAL ESTATE CO, CLARE, IRELAND.

DOCUMENT TYPE: Article; Journal

LANGUAGE: English

REFERENCE COUNT: 26

ENTRY DATE: Entered STN: 2 Feb 2001

Last Updated on STN: 2 Feb 2001

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB 6-Hydroxymellein (6HM) synthase, a multifunctional polyketide biosynthetic enzyme in carrot cells, is capable of catalyzing the acyl-CoA condensation and the ketoreduction in the presence of the nucleotide reducing co-factors. Although free CoA at high concentrations functioned as the activator of the NADPH-dependent 6HM formation, the compound exhibited an appreciable inhibitory activity toward the reaction mediated by NADH. CoA showed a potent inhibitory activity against substrate entry into the reaction center of the NADH-associated enzyme while, in the presence of NADPH, the compound slightly inhibited the formation of the acylated enzyme. The catalytic rate of the synthase was appreciably decreased when NADPH was replaced by the deuterium-labeled compound, however, the k_H/k_D value was markedly reduced if NADH and [D]NADH were employed as the reducing co-factors. These results suggest that the phosphate group attached to 2'-position of the adenosyl moiety of NADPH associated with the ketoreducing domain of 6HM synthase plays an important role in the regulation of the enzyme activity. (C) 2000 Elsevier Science Ireland Ltd. All rights reserved.

L14 ANSWER 14 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1999:593400 HCAPLUS

DOCUMENT NUMBER: 131:347279

TITLE: The complete chloroplast DNA sequence of the green alga *Nephroselmis olivacea*: insights into the architecture of ancestral chloroplast genomes

AUTHOR(S): Turmel, Monique; Otis, Christian; Lemieux, Claude

CORPORATE SOURCE: Canadian Institute for Advanced Research, Program in Evolutionary Biology and Departement de Biochimie, Universite Laval, Quebec, QC, G1K 7P4, Can.

SOURCE: Proceedings of the National Academy of Sciences of the United States of America (1999), 96(18), 10248-10253 CODEN: PNASA6; ISSN: 0027-8424

PUBLISHER: National Academy of Sciences

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Green plants seem to form two sister lineages: Chlorophyta, comprising the green algal classes Prasinophyceae, Ulvophyceae, Trebouxiophyceae, and Chlorophyceae, and Streptophyta, comprising the Charophyceae and land plants. We have determined the complete chloroplast DNA (cpDNA) sequence (200,799 bp) of *Nephroselmis olivacea*, a member of the class (Prasinophyceae) thought to include descendants of the earliest-diverging green algae. The 127 genes identified in this genome represent the largest gene repertoire among the green algal and land plant cpDNAs completely sequenced to date. Of the *Nephroselmis* genes, 2 (*ycf81*, and *ftsI*, a gene involved in peptidoglycan synthesis) have not been identified in any previously investigated cpDNA; 5 genes [*ftsW*, *rnE*, *ycf62*, *rnpB*, and *trnS(cga)*] have been found only in cpDNAs of nongreen algae; and 10 others (*ndh* genes) have been described only in land plant cpDNAs. *Nephroselmis* and land plant cpDNAs share the same quadripartite structure-which is

characterized by the presence of a large rRNA-encoding inverted repeat and two unequal single-copy regions-and very similar sets of genes in corresponding genomic regions. Given that our phylogenetic analyses place *Nephroselmis* within the Chlorophyta, these structural characteristics were most likely present in the cpDNA of the common ancestor of chlorophytes and streptophytes. Comparative analyses of chloroplast genomes indicate that the typical quadripartite architecture and gene-partitioning pattern of land plant cpDNAs are ancient features that may have been derived from the genome of the cyanobacterial progenitor of chloroplasts. Our phylogenetic data also offer insight into the chlorophyte ancestor of euglenophyte chloroplasts.

REFERENCE COUNT: 35 THERE ARE 35 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 15 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 2000:286802 HCAPLUS

DOCUMENT NUMBER: 133:276987

TITLE: The photosynthesis gene cluster of *Rhodobacter sphaeroides*

AUTHOR(S): Naylor, Grant William; Addlesee, Hugh Alistair; Gibson, Lucien Charles Donald; Hunter, Christopher Neil

CORPORATE SOURCE: Robert Hill Institute for Photosynthesis and Krebs Institute for Biomolecular Research, Department of Molecular Biology and Biotechnology, University of Sheffield, Sheffield, UK

SOURCE: Photosynthesis Research (1999), 62(2-3), 121-139
CODEN: PHRSDI; ISSN: 0166-8595

PUBLISHER: Kluwer Academic Publishers

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The photosynthetic bacteria are at the forefront of the study of many aspects of photosynthesis, including photopigment biosynthesis, photosynthetic-membrane assembly, light-harvesting, and reaction center photochem. The facultative growth of some photosynthetic bacteria, their simple photosystems, and their ease of genetic manipulation have all contributed to advances in these areas. Amongst these bacteria, the purple non-sulfur bacterium *Rhodobacter sphaeroides* has emerged as, arguably, the leading contender for a model system in which to integrate the studies of all the different aspects of the assembly and function of the photosynthetic apparatus. Many of the genes encoding photosynthesis-related proteins are known to be clustered within a small region of the genome in this organism. As a further aid to studying the assembly and function of the photosystem of *Rb. sphaeroides*, the DNA sequence for a genomic segment containing this photosynthesis gene cluster (PGC) has been assembled from previous EMBL submissions and formerly unpublished data. The *Rb. sphaeroides* PGC is 40.7 kb in length and consists of 38 open reading frames encoding the reaction center H, L and M subunits, the α and β polypeptides of the light-harvesting I (B875) complex, and the enzymes of bacteriochlorophyll and carotenoid biosynthesis. PGCs are a feature of gene organization in several photosynthetic bacteria, and the similarities between the clusters of *Rb. sphaeroides* and *Rb. capsulatus* have been apparent for some time. Here we present the first comprehensive anal. of the PGC of *Rb. sphaeroides*, as well as a comparison with that of *Rb. capsulatus*.

REFERENCE COUNT: 122 THERE ARE 122 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 16 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1998:805585 HCAPLUS

DOCUMENT NUMBER: 130:180061

TITLE: Tracking molecular evolution of photosynthesis by characterization of a major photosynthesis gene cluster from *Heliobacillus mobilis*

AUTHOR(S): Xiong, Jin; Inoue, Kazuhito; Bauer, Carl E.

CORPORATE SOURCE: Department of Biology, Indiana University, Bloomington, IN, 47405, USA

SOURCE: Proceedings of the National Academy of Sciences of the United States of America (1998), 95(25), 14851-14856
CODEN: PNASA6; ISSN: 0027-8424

PUBLISHER: National Academy of Sciences

DOCUMENT TYPE: Journal

LANGUAGE: English

AB A DNA sequence has been obtained for a 35.6-kb genomic segment from *Helicobacter mobilis* that contains a major cluster of photosynthesis genes. A total of 30 ORFs were identified, 20 of which encode enzymes for bacteriochlorophyll and carotenoid biosynthesis, reaction-center (RC) apoprotein, and cytochromes for cyclic electron transport. Donor side electron-transfer components to the RC include a putative RC-associated cytochrome c553 and a unique four-large-subunit cytochrome bc complex consisting of Rieske Fe-S protein (encoded by *petC*), cytochrome b6 (*petB*), subunit IV (*petD*), and a diheme cytochrome c (*petX*). Phylogenetic anal. of various photosynthesis gene products indicates a consistent grouping of oxygenic lineages that are distinct and descendent from anoxygenic lineages. In addition, *H. mobilis* was placed as the closest relative to cyanobacteria, which form a monophyletic origin to chloroplast-based photosynthetic lineages. The consensus of the photosynthesis gene trees also indicates that purple bacteria are the earliest emerging photosynthetic lineage. Our anal. also indicates that an ancient gene-duplication event giving rise to the paralogous *bchI* and *bchD* genes predates the divergence of all photosynthetic groups. In addition, our anal. of gene duplication of the photosystem I and photosystem II core polypeptides supports a "heterologous fusion model" for the origin and evolution of oxygenic photosynthesis.

REFERENCE COUNT: 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L14 ANSWER 17 OF 19 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1995:730378 HCAPLUS

DOCUMENT NUMBER: 123:136396

TITLE: Abundant PLPP sequence has the same conformation in unrelated proteins

AUTHOR(S): Zielenkiewicz, Piotr; Plochocka, Danuta

CORPORATE SOURCE: Inst. Biochemistry Biophysics, Polish Academy Sci., Warsaw, 02-106, Pol.

SOURCE: Protein and Peptide Letters (1995), 2(1), 299-304
CODEN: PPELEN; ISSN: 0929-8665

PUBLISHER: Bentham Science Publishers BV

DOCUMENT TYPE: Journal

LANGUAGE: English

AB It has been shown that a low complexity sequence PLPP is abundantly present in a representative set of PIR database. The PLPP fragment has the same loop conformation in five unrelated protein structures. Conformational anal. of PXPP tetrapeptides, as well as anal. of known protein structures, shows that this particular conformation should be adopted for an X more bulky than valine.

L14 ANSWER 18 OF 19 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN DUPLICATE 1

ACCESSION NUMBER: 1992:9786 BIOSIS

DOCUMENT NUMBER: PREV199293009786; BA93:9786

TITLE: REGULATION OF CHLOROPLAST METABOLISM IN LEAVES EVIDENCE THAT NADP-DEPENDENT GLYCERALDEHYDEPHOSPHATE DEHYDROGENASE BUT NOT FERREDOXIN-NADP REDUCTASE CONTROLS ELECTRON FLOW TO PHOSPHOGLYCERATE IN THE DARK-LIGHT TRANSITION.

AUTHOR(S): SIEBKE K [Reprint author]; LAISK A; NEIMANIS S; HEBER U

CORPORATE SOURCE: INSTITUTE BOTANY PHARMACEUTICAL BIOLOGY, UNIVERSITY WUERZBURG, W-8700 WUERZBURG, W GER

SOURCE: Planta (Heidelberg), (1991) Vol. 185, No. 3, pp. 337-343.
CODEN: PLANAB. ISSN: 0032-0935.

DOCUMENT TYPE: Article

FILE SEGMENT: BA

LANGUAGE: ENGLISH

ENTRY DATE: Entered STN: 10 Dec 1991
Last Updated on STN: 6 Mar 1992

AB P700 is rapidly, but only transiently photooxidized upon illuminating dark-adapted leaves. Initial oxidation is followed by a reductive phase even under far-red illumination which excites predominantly photosystem (PS) I. In this phase, oxidized P700 is reduced by electrons coming from PSII. Charge separation in the reaction center of PSI is prevented by the unavailability of electron acceptors on the reducing side of PSI. It is subsequently made possible by the opening of an electron gate which is situated between PSI and the electron acceptor phosphoglycerate. Electron acceptors immediately available for reduction while the gate is closed corresponded to 10 nmol · (mg chlorophyll)⁻¹ electrons in geranium leaves, 16 nmol · (mg chlorophyll)⁻¹ in sunflower and 22 nmol · (mg chlorophyll)⁻¹ in oleander. Reduction of NADP during the initial phase of P700 oxidation showed that the electron gate was not represented by ferredoxin-NADP reductase. Availability of ATP indicated that electron flow was not hindered by deactivation of the thylakoid ATP synthetase. It is concluded that NADP-dependent glyceraldehydephosphate dehydrogenase is completely deactivated in the dark and activated in the light. The rate of activation depends on the length of the preceding dark period. As chloroplasts contain both NAD- and NADP-dependent glyceraldehydephosphate dehydrogenases, deactivation of the NADP-dependent enzyme disconnects chloroplast NAD and NADP systems and prevents phosphoglycerate reduction in the dark at the expense of NADPH and ATP which are generated by glucose-6-phosphate oxidation and glycolytic starch breakdown, respectively.

L14 ANSWER 19 OF 19 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1980:226910 BIOSIS
DOCUMENT NUMBER: PREV198070019406; BA70:19406
TITLE: PHYSIOLOGY OF PHOTOSYNTHESIS IN HIGHER PLANTS THE ADAPTATION TO LIGHT INTENSITY AND LIGHT QUALITY.
AUTHOR(S): WILD A [Reprint author]
CORPORATE SOURCE: INST ALLG BOT, UNIV MAINZ, SAARSTR 21, D-6500 MAINZ, W GER
SOURCE: Berichte der Deutschen Botanischen Gesellschaft, (1980) Vol. 92, No. 2-3, pp. 341-364.
CODEN: BEDBAP. ISSN: 0365-9631.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: GERMAN

AB Rapid progress has been made in the study of the mechanisms of photosynthesis in the past 30 yr. It is now possible to study in detail the component steps of the highly complex overall process. The knowledge of the basic mechanisms of the photosynthetic process provided the base for a renaissance of modern physiological studies. A comparative study is given on the effect of light intensity and light quality concerning photosynthetic adaptation of higher plants during growth. Low-light and high-light plants differ in a number of component steps of photosynthesis as well as in the structure and composition of the photosynthetic apparatus and in the leaf anatomy. Photosynthetic adaptation to different light levels involves balanced changes of many leaf factors so that not only one single reaction limits light-saturated rates of photosynthesis under natural conditions. Special attention is paid to the changes of the structure of the thylakoid membranes and to the variability of the size of the photosynthetic unit. In low-light plants the capacity of the pigment system is increased whereas there is a decrease in the concentration of the redox enzymes of the electron transport chain. In the high-light plants the experimental data can be well explained by the model of the tripartite organization of the photosynthetic unit. In low-light plants a multipartite organization has to be postulated. Results indicate that an increase in the size of the photosynthetic unit is not realized by establishing a higher ratio of chlorophyll per reaction center but by the co-operation of several antennae systems. In the partition regions of the grana several antennae systems I, antennae systems II and light-harvesting complexes can communicate with 1 electron transport chain and therefore belong to one photosynthetic unit. Even if not all P-700 and P-680 were involved directly in the electron transport chain at least their chlorophyll complexes should be so connected that resonant energy transfer to distinct reaction center

could be possible. The model demonstrates a P-700/cytochrome f (cyt f) f ratio of 2:1. By transferring it into three dimensions, the model will provide enough space for even bigger photosynthetic units with a ratio of P-700/Cyt f of 3:1 to 4:1. The Chl(chlorophyll)/Cyt f ratio is used to estimate the size of the photosynthetic unit. On the other hand the ratio of Chl/P-700 can not be a measure for the size of the photosynthetic unit in weak-light plants as shown by the multipartite model. Such a definition would have no physiological relevance in low-light plants. The partition regions in the grana are pigment-rich membranes. Partition formation during the development of chloroplasts is correlated with the synthesis of LH (light harvesting) Chl. The synthesis of LH Chl leads to an increase in the size of the photosynthetic unit. In plants, which have to grow in low-light, the limiting factor for photosynthesis is light. Thus, these plants need high pigment concentrations and low enzyme contents. The grana stacks in low-light plants provide the possibility for the co-operation of several antennae systems for the collection of light quanta. This is advantageous, as it allows a more efficient collection of light per electron transport chain. The discrepancy between the absorption power of chlorophyll and the rate capacity of the electron transport system is reduced by the larger size of the photosynthetic unit in low-light plants. An increase in the size of the photosynthetic unit leads to a decrease in the light energy needed to saturate photosynthesis and to a decrease in the photosynthetic rate based on chlorophyll content. Therefore, the differences in the light required for saturation and the light-saturated rate of photosynthesis under high external CO₂-concentration are explained by the variability of the size of the photosynthetic unit. The outer membranes of the grana and the stroma thylakoids contain PS I, NADP⁺-reductase and ATP-synthetase. NADP⁺-reduction and ATP-synthesis only occur at the surface of the o.

=> d his

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

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L1      4 S ASTAXANTHIN (W)SYNTHASE?
L2      0 S ASTAXANTHIN (W)SYNTHETASE?
L3      6416 S ASTAXANTHIN
L4      24046 S "AST"
L5      194127 S SYNTHETASE?
L6      0 S L3 (W)L5
L7      39 S L3 AND L5
L8      24 DUP REM L7 (15 DUPLICATES REMOVED)
L9      41050 S REACTION (A) CENTER
L10     0 S L8 AND L9
L11     153 S L5 AND L9
L12     371127 S REDUCTASE?
L13     21 S L11 AND L12
L14     19 DUP REM L13 (2 DUPLICATES REMOVED)
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=> s P450 reductase?

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L15     7593 P450 REDUCTASE?
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=> s l9 and l15

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L16     0 L9 AND L15
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=> s l4 and l9

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L17     0 L4 AND L9
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=> d his

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

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L1      4 S ASTAXANTHIN (W)SYNTHASE?
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L2 0 S ASTAXANTHIN (W) SYNTHETASE?
 L3 6416 S ASTAXANTHIN
 L4 24046 S "AST"
 L5 194127 S SYNTHETASE?
 L6 0 S L3 (W) L5
 L7 39 S L3 AND L5
 L8 24 DUP REM L7 (15 DUPLICATES REMOVED)
 L9 41050 S REACTION (A) CENTER
 L10 0 S L8 AND L9
 L11 153 S L5 AND L9
 L12 371127 S REDUCTASE?
 L13 21 S L11 AND L12
 L14 19 DUP REM L13 (2 DUPLICATES REMOVED)
 L15 7593 S P450 REDUCTASE?
 L16 0 S L9 AND L15
 L17 0 S L4 AND L9

=> s "beta-carotene"

L18 59612 "BETA-CAROTENE"

=> s l4 and l18

L19 64 L4 AND L18

=> s l9 and l19

L20 0 L9 AND L19

=> l1 and l9

L1 IS NOT A RECOGNIZED COMMAND

The previous command name entered was not recognized by the system.

For a list of commands available to you in the current file, enter

"HELP COMMANDS" at an arrow prompt (=>).

=> s l1 and l9

L21 0 L1 AND L9

=> dup rem l1

PROCESSING COMPLETED FOR L1

L22 3 DUP REM L1 (1 DUPLICATE REMOVED)

=> d 1-3 ibib ab

L22 ANSWER 1 OF 3 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 2005-27515 BIOTECHDS

TITLE: Stably expressing a nucleic acid molecule in one carbon
 metabolizing microorganism comprises integrating nucleic acid
 molecule in tig region of genome of microorganism;
 vector-mediated gene transfer and expression in host cell
 for strain improvement and carotenoid compound production

AUTHOR: MILLER E S; YE R W

PATENT ASSIGNEE: DU PONT DE NEMOURS and CO E I

PATENT INFO: WO 2005087942 22 Sep 2005

APPLICATION INFO: WO 2005-US7120 4 Mar 2005

PRIORITY INFO: US 2004-550385 5 Mar 2004; US 2004-550385 5 Mar 2004

DOCUMENT TYPE: Patent

LANGUAGE: English

OTHER SOURCE: WPI: 2005-658903 [67]

AB DERWENT ABSTRACT:

NOVELTY - A nucleic acid molecule is stably expressed in a one carbon
 metabolizing microorganism by providing a one carbon metabolizing
 microorganism having a tig region in the genome; integrating nucleic acid
 molecule(s) into the tig region of the genome of the metabolizing
 microorganism; and growing the metabolizing microorganism under
 conditions where the nucleic acid molecule is stably-expressed, is new.

DETAILED DESCRIPTION - INDEPENDENT CLAIMS are also included for: (A)
 production of a carotenoid compound comprising providing one carbon (1C)
 metabolizing microorganism comprising a gene cluster comprising genes
 encoding the carotenoid biosynthetic pathway inserted into the tig region
 of the genome; contacting the metabolizing microorganism with 1C carbon
 substrate from methane and/or methanol under conditions where the gene

cluster is expressed and a carotenoid compound is produced; and optionally recovering the carotenoid compound; (B) a 1C metabolizing microorganism comprising a nucleic acid molecule integrated in the tig region of the genome; and (C) identifying an integration site in a genome for high level expression of a nucleic acid molecule microorganism comprising: (a) providing an integration vector comprising a gene cluster encoding the following enzymes: geranylgeranyl pyrophosphate synthase, zeaxanthin glucosyl transferase; lycopene cyclase, phytoene desaturase, phytoene synthase, 13-carotene hydroxylase, 13-carotene ketolase, and isopentenyl diphosphate isomerase, to facilitate the integration of the gene cluster in to the genome of a microorganism; (b) contacting the integration vector with a microorganism under conditions that allow for random integration of the gene cluster into the microorganism genome to create random transformants; (c) screening the random transformants for expression of the gene cluster on the basis of the production of a 40C carotenoid; and (d) identifying sites of integration of the gene cluster into the genome of the random transformants.

BIOTECHNOLOGY - Preferred Method: The nucleic acid molecule is transcribed using the tig promoter. It is operably integrated. Multiple unlinked genes are integrated at different positions within the tig region. The nucleic acid molecule is integrated into the tig region downstream of the tig promoter or of any gene of the tig region. It can be integrated downstream of the tig open or ion open reading frame, or downstream of the clpP open reading frame. **Preferred Nucleic Acid:** The nucleic acid molecule lacks an antibiotic selection marker. The nucleic acid molecule comprises multiple tandem genes in a single fragment. It is a gene. The tig region is defined according to a fully defined 9010 base pairs sequence given in the specification. The nucleic acid molecule is genes encoding transaldolase, fructose biphosphate aldolase, keto deoxy phosphogluconate aldolase, phosphoglucomutase, glucose-6-phosphate isomerase, phosphofructokinase, 6-phosphogluconate dehydratase, 6-phosphogluconate-6-phosphate-1 dehydrogenase, dxs, dxr, ispA, ispD, ispE, ispF, crtE, crtX, crtY, crtI, crtB, crtZ, crtD, crtO, crtW, crtidi, genes encoding limonene synthase, ugp, gumD, wza, espB, espM, waaE, espV, gumH, genes encoding glycosyltransferase genes, aroG, aroB, aroQ, aroE, aroK, 5-enolpyruvylshikimate-3phosphate synthase, aroC, trpE, trpD, trpC, trpB, pheA, tyrAc, pds, phaC, phaE, efe, pdc, adh, pinene synthase, bornyl synthase, phellandrene synthase, cineole synthase, sabinene synthase, or taxadiene synthase. It encodes at least one enzyme in the carotenoid biosynthetic pathway. The enzyme in the carotenoid biosynthetic pathway is geranylgeranyl pyrophosphate synthase, zeaxanthin glucosyl transferase, lycopene cyclase, phytoene desaturase, phytoene synthase, beta-carotene hydroxylase, beta-carotene ketolase, or isopentenyl diphosphate isomerase. **Preferred Microorganism:** The metabolizing microorganism is methanotrophs or methylotrophs. It can be Methylomonas, Methylobacter, Methylococcus, Methylosinus, Methylocystis, Methylochromium, Methanomonas, Methylophilus, Methylobacillus, Methylobacterium, Hyphomicrobium, Xanthobacter, Bacillus, Paracoccus, Nocardia, Arthrobacter, Rhodopseudomonas, or Pseudomonas. It is Methylomonas 16a. The metabolizing microorganism has the ATCC designation ATCC PTA 2402. **Preferred Carotenoid:** The carotenoid compound is. antheraxanthin, adonixanthin, astaxanthin, canthaxanthin, aanthaxanthin, capsorubrin, alpha-cryptoxanthin alpha-carotene, beta-carotene, epsilon-carotene, echinenone, gamma-carotene, zeta-carotene, alpha-cryptoxanthin, diatoxanthin, 7,8-didehydroastaxanthin, fucoxanthin, fucoxanthinol, isorenieratene, lactucaxanthin, lutein, lycopene, neoxanthin, neurosporene, hydroxyneurosporene, peridinin, phytoene, rhodopin, rhodopin glucoside, siphonaxanthin, spheroidene, spheroidenone, spirilloxanthin, uriolide, uriolide acetate, violaxanthin, zeaxanthin-beta-diglucoside, zeaxanthin, or canthaxanthin.

USE - For stably expressing a nucleic acid molecule in 1C metabolizing microorganism, useful for production of a carotenoid compound (claimed).

ADVANTAGE - The invention results in high level and stable production of 40C carotenoids.

EXAMPLE - The growth of Methylomonas sp. 16a for tri-parental mating initiated with the inoculation of -80degreesC frozen stock culture into 20 ml ammonium liquid medium containing 25% methane. The culture was grown at 30degreesC with aeration. This saturated culture was used to

inoculate 100 ml of fresh ammonium liquid medium containing 25% methane. The 100 ml culture was grown at 30degreesC with aeration until the culture reached an OD600 between 0.7-0.8. The Methylomonas cell pellets were re-suspended. The re-suspended Methylomonas cells were used to re-suspend the combined Escherichia coli and helper cell pellets. The cell suspension was spotted on agar plates containing 0.05% yeast extract. The plates were incubated at 30degreesC for 3 days in a jar containing 25% methane. The cells from the cultures were inoculated in ammonium liquid medium and 25% methane and grown overnight at 30degreesC with aeration. The cultures were monitored for Escherichia coli growth by plating on agar plates to verify the elimination of the Escherichia coli. (136 pages)

L22 ANSWER 2 OF 3 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 2002:281124 BIOSIS
DOCUMENT NUMBER: PREV200200281124
TITLE: **Astaxanthin synthase.**
AUTHOR(S): Hoshino, Tatsuo [Inventor, Reprint author]; Ojima, Kazuyuki [Inventor]; Setoguchi, Yutaka [Inventor]
CORPORATE SOURCE: Kamakura, Japan
ASSIGNEE: Roche Vitamins Inc.
PATENT INFORMATION: US 6365386 20020402
SOURCE: Official Gazette of the United States Patent and Trademark Office Patents, (Apr. 2, 2002) Vol. 1257, No. 1.
<http://www.uspto.gov/web/menu/patdata.html>. e-file.
CODEN: OGUPE7. ISSN: 0098-1133.
DOCUMENT TYPE: Patent
LANGUAGE: English
ENTRY DATE: Entered STN: 8 May 2002
Last Updated on STN: 25 Jun 2002

AB The present invention is directed to genetic materials useful for the preparation of astaxanthin from beta-carotene, such as polypeptides having **astaxanthin synthase** activity, DNA fragments coding for **astaxanthin synthase**, recombinant organisms and the like. Those novel genetic materials may be derived from *Phaffia rhodozyma*. The present invention also provides a process for the production of astaxanthin.

L22 ANSWER 3 OF 3 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN
DUPLICATE 1

ACCESSION NUMBER: 2000-14326 BIOTECHDS
TITLE: Novel polynucleotide encoding **astaxanthin-synthase** useful for producing recombinant cells for producing astaxanthin from beta-carotene;
plasmid pRLR913-mediated gene transfer and expression in *Phaffia rhodozyma*
AUTHOR: Hoshino T; Ojima K; Setoguchi Y
PATENT ASSIGNEE: Roche
LOCATION: Basle, Switzerland.
PATENT INFO: EP 1035206 13 Sep 2000
APPLICATION INFO: EP 2000-104430 3 Mar 2000
PRIORITY INFO: EP 20002101666 1 Feb 2000; EP 1999-104668 9 Mar 1999
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 2000-559874 [52]

AB A N containing nucleotide sequence coding for an enzyme with **astaxanthin-synthase** activity catalyzing the reaction from beta-carotene to astaxanthin, preferably in *Phaffia rhodozyma*, is new. Also claimed are: a vector or plasmid (e.g. plasmid pRLR913) containing the DNA; a host cell (e.g. *P. rhodozyma* (ATCC 96815)) transformed or transfected by the DNA or vector; a protein encoded by the DNA; preparation of the protein by culturing the host cell under suitable conditions and recovering the expressed protein; and production of astaxanthin by contacting beta-carotene with a peptide with **astaxanthin-synthase** activity in the presence of an electron donor in a reaction mixture containing a reconstituted membrane. The DNA and protein are useful for producing astaxanthin. (46pp)

=> d 11 1-3 kwic

L1 ANSWER 1 OF 4 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
TI **Astaxanthin synthase.**
AB. . . The present invention is directed to genetic materials useful for
the preparation of astaxanthin from beta-carotene, such as polypeptides
having **astaxanthin synthase** activity, DNA fragments
coding for **astaxanthin synthase**, recombinant organisms
and the like. Those novel genetic materials may be derived from *Phaffia*
rhodozyma. The present invention also provides. . .
IT Major Concepts
Molecular Genetics (Biochemistry and Molecular Biophysics);
Pharmacology
IT Chemicals & Biochemicals
DNA fragments; astaxanthin: biosynthesis, therapeutic potential;
astaxanthin synthase; beta-carotene
RN 472-61-7 (astaxanthin)
7235-40-7 (beta-carotene)
293749-46-9 (**ASTAXANTHIN SYNTHASE**)

L1 ANSWER 2 OF 4 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN
CT . . . ESPB, ESPM, WAAE, ESPV, GUMH GLYCOSYLTRANSFERASE, AROG, AROB,
AROQ, AROE, AROK 5-ENOLPYRUVYLSHIKIMATE-3-PHOSPHATE-SYNTHASE, AROC, TRPE,
TRPD, TRPC, TRPB, PHEA, TYRAC, PDS, PHAC, PHAE, EFE, PDC, ADH,
PINENE-SYNTHASE, BORNYL-SYNTHASE, PHELLANDRENE-SYNTHASE,
CINEOLE-SYNTHASE, SABINENE-SYNTHASE, TAXADIENE-SYNTHASE GENE TRANSFER,
EXPRESSION IN HOST CELL, TIG REGION GENOME. . .

L1 ANSWER 3 OF 4 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN
TI Novel polynucleotide encoding **astaxanthin-synthase**
useful for producing recombinant cells for producing astaxanthin from
beta-carotene;
plasmid pRLR913-mediated gene transfer and expression in *Phaffia*
rhodozyma
AB A N containing nucleotide sequence coding for an enzyme with
astaxanthin-synthase activity catalyzing the reaction
from beta-carotene to astaxanthin, preferably in *Phaffia rhodozyma*, is
new. Also claimed are: a vector or. . . cell under suitable
conditions and recovering the expressed protein; and production of
astaxanthin by contacting beta-carotene with a peptide with
astaxanthin-synthase activity in the presence of an
electron donor in a reaction mixture containing a reconstituted membrane.
The DNA and protein. . .
CT PLASMID PRLR913-MEDIATED **ASTAXANTHIN-SYNTHASE** GENE
TRANSFER, EXPRESSION IN *PHAFFIA RHODOZYMA*, BETA-CAROTENE CONVERSION APPL.
ASTAXANTHIN PREP., ANTIOXIDANT PIGMENT CAROTENOID KETONE ALCOHOL YEAST
FUNGUS (VOL.19, NO.25)

=> d his

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,
LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

L1 4 S ASTAXANTHIN (W) SYNTHASE?
L2 0 S ASTAXANTHIN (W) SYNTHETASE?
L3 6416 S ASTAXANTHIN
L4 24046 S "AST"
L5 194127 S SYNTHETASE?
L6 0 S L3 (W) L5
L7 39 S L3 AND L5
L8 24 DUP REM L7 (15 DUPLICATES REMOVED)
L9 41050 S REACTION (A) CENTER
L10 0 S L8 AND L9
L11 153 S L5 AND L9
L12 371127 S REDUCTASE?
L13 21 S L11 AND L12
L14 19 DUP REM L13 (2 DUPLICATES REMOVED)

L15 7593 S P450 REDUCTASE?
 L16 0 S L9 AND L15
 L17 0 S L4 AND L9
 L18 59612 S "BETA-CAROTENE"
 L19 64 S L4 AND L18
 L20 0 S L9 AND L19
 L21 0 S L1 AND L9
 L22 3 DUP REM L1 (1 DUPLICATE REMOVED)

=> dup rem l11

PROCESSING COMPLETED FOR L11

L23 93 DUP REM L11 (60 DUPLICATES REMOVED)

=> d 80-93 ibib ab

L23 ANSWER 80 OF 93 MEDLINE on STN
 ACCESSION NUMBER: 86165318 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 3007236
 TITLE: Chloroplast genes for photosynthetic membrane components of higher plants.
 AUTHOR: Gray J C; Bird C R; Courtice G R; Hird S M; Howe C J; Huttly A K; Phillips A L; Smith A G; Willey D L; Bowman C M; +
 SOURCE: Biochemical Society transactions, (1986 Feb) 14 (1) 25-7. Journal code: 7506897. ISSN: 0300-5127.
 PUB. COUNTRY: ENGLAND: United Kingdom
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 198605
 ENTRY DATE: Entered STN: 19900321
 Last Updated on STN: 19900321
 Entered Medline: 19860520

L23 ANSWER 81 OF 93 MEDLINE on STN DUPLICATE 21
 ACCESSION NUMBER: 85252766 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 4016097
 TITLE: Gramicidin S **synthetase**. Temperature dependence and thermodynamic parameters of substrate amino acid activation reactions.
 AUTHOR: Vater J; Mallow N; Gerhardt S; Gadow A; Kleinkauf H
 SOURCE: Biochemistry, (1985 Apr 9) 24 (8) 2022-7. Journal code: 0370623. ISSN: 0006-2960.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 198509
 ENTRY DATE: Entered STN: 19900320
 Last Updated on STN: 19980206
 Entered Medline: 19850916

AB In the biosynthesis of the cyclic decapeptide antibiotic gramicidin S, the constituent amino acids are activated by a two-step mechanism involving aminoacyl adenylate and thio ester formation which are both reversible processes. The dissociation constants (KD) for the gramicidin S **synthetase**-substrate amino acid-thio ester complexes are 100-1000-fold lower compared to the KM data of the preceding aminoacyl adenylate reactions. The affinity for these substrates is appreciably higher at the thio template sites than at the aminoacyl adenylate **reaction centers**. Therefore, the activation equilibria are quantitatively shifted toward thio ester formation. A set of thermodynamic parameters for the activation processes was determined from the temperature dependence of the KM and KD data. Reaction enthalpies were obtained from a van't Hoff analysis of these constants. delta G degree for the substrate activation reactions of the heavy enzyme of gramicidin S **synthetase** (GS 2) is predominantly controlled by entropy contributions. In contrast, the overall activation and concomitant racemization of phenylalanine by phenylalanine racemase (GS 1) are exothermic processes which are distinguished by a small negative

reaction entropy.

L23 ANSWER 82 OF 93 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1985:57863 HCAPLUS

DOCUMENT NUMBER: 102:57863

TITLE: From bioenergetics to membrane protein structure and gene expression

AUTHOR(S): Ferguson, S. J.

CORPORATE SOURCE: Dep. Biochem., Univ. Birmingham, Birmingham, B15 2TT, UK

SOURCE: Trends in Biochemical Sciences (1984), 9(12), 501-2

CODEN: TBSCDB; ISSN: 0376-5067

DOCUMENT TYPE: Journal; General Review

LANGUAGE: English

AB A review with 2 refs., of the crystal structure of the photosynthetic reaction center of Rhodospseudomonas viridis and on the basis for enhanced expression of a particular cistron of a polycistronic mRNA that contains coding information for the 8 subunits of Escherichia coli ATP synthase.

L23 ANSWER 83 OF 93 MEDLINE on STN

ACCESSION NUMBER: 84240031 MEDLINE

DOCUMENT NUMBER: PubMed ID: 6330136

TITLE: Correlation of structure and function of chloroplast membranes at the supramolecular level.

AUTHOR: Staehelin L A; DeWit M

CONTRACT NUMBER: GM 18639 (NIGMS)

GM 22912 (NIGMS)

SOURCE: Journal of cellular biochemistry, (1984) 24 (3) 261-9.

Journal code: 8205768. ISSN: 0730-2312.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 198408

ENTRY DATE: Entered STN: 19900320

Last Updated on STN: 19970203

Entered Medline: 19840813

AB Freeze-fracture electron microscopy has revealed that different size classes of intramembrane particles of chloroplast membranes are nonrandomly distributed between appressed grana and nonappressed stroma membrane regions. It is now generally assumed that thylakoid membranes contain five major functional complexes, each of which can give rise to an intramembrane particle of a defined size. These are the photosystem II complex, the photosystem I complex, the cytochrome f/b6 complex, the chlorophyll a/b light-harvesting complex, and the CF0 -CF1 ATP synthetase complex. By mapping the distribution of the different categories of intramembrane particles, information on the lateral organization of functional membrane units of thylakoid membranes can be determined. In this review, we present a brief summary of the evidence supporting the correlation of specific categories of intramembrane particles with known biochemical entities. In addition, we discuss studies showing that ions and phosphorylation of the membrane adhesion factor, the chlorophyll a/b light-harvesting complex, can affect the lateral organization of chloroplast membrane components and thereby regulate membrane function.

L23 ANSWER 84 OF 93 MEDLINE on STN

DUPLICATE 22

ACCESSION NUMBER: 83182451 MEDLINE

DOCUMENT NUMBER: PubMed ID: 6188612

TITLE: Gramicidin S synthetase. Stability of reactive thioester intermediates and formation of 3-amino-2-piperidone.

AUTHOR: Gadow A; Vater J; Schlumbohm W; Palacz Z; Salnikow J; Kleinkauf H

SOURCE: European journal of biochemistry / FEBS, (1983 May 2) 132 (2) 229-34.

Journal code: 0107600. ISSN: 0014-2956.

PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198306
ENTRY DATE: Entered STN: 19900318
Last Updated on STN: 19980206
Entered Medline: 19830610

AB The reactive thioester complexes of gramicidin S synthetase with substrate amino acids and intermediate peptides are slowly hydrolyzed in neutral buffer solutions under mild conditions. Fully active enzyme is recovered. These processes are strongly accelerated by certain thiol protective agents. In the presence of 1 mM dithioerythritol the half-life times of these hydrolysis reactions are in the range of 1-90 h at 3 degrees C. The thioester complex of gramicidin S synthetase 2 (GS2, the heavy enzyme) with the tripeptide DPhe-Pro-Val is distinguished by the highest stability of all these intermediates. A different decomposition pattern is observed for the thioester complex of GS2 with LOrn. Here 3-amino-2-piperidone (cyclo-LOrn) is formed in a rapid cyclization reaction. This product specifically blocks the activation center of GS2 for LOrn at the thioester binding site. All other activation reactions of gramicidin S synthetase are unaffected. A procedure for a specific labelling of the reaction centers of the multienzyme is outlined.

L23 ANSWER 85 OF 93 MEDLINE on STN
ACCESSION NUMBER: 83012558 MEDLINE
DOCUMENT NUMBER: PubMed ID: 6750355
TITLE: The role of chlorophyll-protein complexes in the function and structure of chloroplast thylakoids.
AUTHOR: Anderson J M
SOURCE: Molecular and cellular biochemistry, (1982 Aug 6) 46 (3) 161-72.
Journal code: 0364456. ISSN: 0300-8177.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198212
ENTRY DATE: Entered STN: 19900317
Last Updated on STN: 19900317
Entered Medline: 19821202

AB The photosynthetic pigments of chloroplast thylakoid membranes are complexed with specific intrinsic polypeptides which are included in three supramolecular complexes, photosystem I complex, photosystem II complex and the light-harvesting complex. There is marked lateral heterogeneity in the distribution of these complexes along the membrane with photosystem II complex and its associated light-harvesting complex being located mainly in the stacked membranes of the grana partitions, while photosystem I complex is found mainly in unstacked thylakoids together with ATP synthetase. In contrast, the intermediate electron transport complex, the cytochrome b-f complex, is rather uniformly distributed in these two membrane regions. The consequences of this lateral heterogeneity in the location of the thylakoid complexes are considered in relation to the function and structure of chloroplasts of higher plants.

L23 ANSWER 86 OF 93 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN DUPLICATE 23
ACCESSION NUMBER: 1983:155612 BIOSIS
DOCUMENT NUMBER: PREV198375005612; BA75:5612
TITLE: THE LOCALIZED COUPLING OF BACTERIAL PHOTO PHOSPHORYLATION EFFECT OF ANTIMYCIN A AND N N DI CYCLOHEXYL CARBODIIMIDE IN CHROMATOPHORES FROM RHODOPSEUDOMONAS-SPHAEROIDES GA STUDIED BY SINGLE TURNOVER EVENT ANALYSIS.
AUTHOR(S): VENTUROLI G [Reprint author]; MELANDRI B A
CORPORATE SOURCE: INST OF BOTANY, UNIV OF BOLOGNA, VIA IRNERIO 42, 40126 BOLOGNA, ITALY
SOURCE: Biochimica et Biophysica Acta, (1982) Vol. 680, No. 1, pp. 8-16.
CODEN: BBACAQ. ISSN: 0006-3002.

DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH

AB The inhibition by antimycin A of the cyclic electron transfer was studied in chromatophores from *R. sphaeroides* Ga following an approach based on the analysis of the relaxation kinetics of the reaction center optical changes in pulsed light. The recovery kinetics of the bacteriochlorophyll redox state are clearly biphasic. The half-times of the fast phase (13 ms) and slow phase (about 400 ms) were not modified by antimycin in a range of concentrations from 0.1-9 μ M. The percentage extent of the fast phase, which reflects the rate of the cyclic electron transfer, was monotonically decreased by increasing concentrations of the inhibitor. This indicates that antimycin decreases progressively the fraction of the photosynthetic units, active in cyclic electron transfer. The ATP yield per flash observed under conditions of controlled inhibition of electron flow was strongly dependent upon the amount of active redox cycles. The amplitude of the carotenoid band shift, which was demonstrated unequivocally to be correlated to the ATP yield per flash in uninhibited chromatophores, was not affected by antimycin up to a 40% inhibition of electron flow. The effect of a progressive limitation by DCCD [dicyclohexylcarbodiimide] in the number of active ATP synthetase complexes on flash-induced phosphorylation was examined. The decrease in ATP yield observed over a wide range of flash frequencies is related simply to the ATPase activity and to phosphorylation in continuous light, irrespective of the value of the membrane potential, which appears to be stabilized by this inhibitor. As a whole, the results obtained at low concentrations of antimycin and under conditions of partial inhibition by DCCD evidence a localized coupling between the redox reactions and phosphorylation.

L23 ANSWER 87 OF 93 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on
STN DUPLICATE 24

ACCESSION NUMBER: 1981:263192 BIOSIS
DOCUMENT NUMBER: PREV198172048176; BA72:48176
TITLE: CHARACTERIZATION OF GLUTAMINE SYNTHETASE FROM
BETA-VULGARIS CULTIVAR ROTA.
AUTHOR(S): NESSELHUT T [Reprint author]; HARNISCHFEGER G
CORPORATE SOURCE: LEHRSTUHL BIOCHEMIE PFLANZE, UNIV GOETTINGEN, 3400
GOETTINGEN, GERMANY
SOURCE: Physiologia Plantarum, (1981) Vol. 51, No. 4, pp. 329-334.
CODEN: PHPLAI. ISSN: 0031-9317.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH

AB Glutamine-synthetase (GS) from *B. vulgaris* seedlings, purified 150-fold, was characterized with regard to its physiologically substrate NH_3 . The data were compared to the unphysiological substrate $\text{NH}_2\text{-OH}$ frequently used in the assay (both synthetase and transferase reaction). The pH-optimum was at pH 7.5 for the synthetase and at pH 6.3 for the transferase reaction. Through plots of pK_m vs. pH, the pK_e values for dissociable groups in the reaction center were in the range from pH 7-8. Mg^{2+} -ion serves as an allosteric effector with a Hill coefficient of 4.2. The results are discussed in relation to the control of N metabolism in Beta.

L23 ANSWER 88 OF 93 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on
STN

ACCESSION NUMBER: 1980:226910 BIOSIS
DOCUMENT NUMBER: PREV198070019406; BA70:19406
TITLE: PHYSIOLOGY OF PHOTOSYNTHESIS IN HIGHER PLANTS THE
ADAPTATION TO LIGHT INTENSITY AND LIGHT QUALITY.
AUTHOR(S): WILD A [Reprint author]
CORPORATE SOURCE: INST ALLG BOT, UNIV MAINZ, SAARSTR 21, D-6500 MAINZ, W GER
SOURCE: Berichte der Deutschen Botanischen Gesellschaft, (1980)
Vol. 92, No. 2-3, pp. 341-364.
CODEN: BEDBAP. ISSN: 0365-9631.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: GERMAN

AB Rapid progress has been made in the study of the mechanisms of photosynthesis in the past 30 yr. It is now possible to study in detail the component steps of the highly complex overall process. The knowledge of the basic mechanisms of the photosynthetic process provided the base for a renaissance of modern physiological studies. A comparative study is given on the effect of light intensity and light quality concerning photosynthetic adaptation of higher plants during growth. Low-light and high-light plants differ in a number of component steps of photosynthesis as well as in the structure and composition of the photosynthetic apparatus and in the leaf anatomy. Photosynthetic adaptation to different light levels involves balanced changes of many leaf factors so that not only one single reaction limits light-saturated rates of photosynthesis under natural conditions. Special attention is paid to the changes of the structure of the thylakoid membranes and to the variability of the size of the photosynthetic unit. In low-light plants the capacity of the pigment system is increased whereas there is a decrease in the concentration of the redox enzymes of the electron transport chain. In the high-light plants the experimental data can be well explained by the model of the tripartite organization of the photosynthetic unit. In low-light plants a multipartite organization has to be postulated. Results indicate that an increase in the size of the photosynthetic unit is not realized by establishing a higher ratio of chlorophyll per reaction center but by the co-operation of several antennae systems. In the partition regions of the grana several antennae systems I, antennae systems II and light-harvesting complexes can communicate with 1 electron transport chain and therefore belong to one photosynthetic unit. Even if not all P-700 and P-680 were involved directly in the electron transport chain at least their chlorophyll complexes should be so connected that resonant energy transfer to distinct reaction center could be possible. The model demonstrates a P-700/cytochrome f (cyt f) f ratio of 2:1. By transferring it into three dimensions, the model will provide enough space for even bigger photosynthetic units with a ratio of P-700/Cyt f of 3:1 to 4:1. The Chl(chlorophyll)/Cyt f ratio is used to estimate the size of the photosynthetic unit. On the other hand the ratio of Chl/P-700 can not be a measure for the size of the photosynthetic unit in weak-light plants as shown by the multipartite model. Such a definition would have no physiological relevance in low-light plants. The partition regions in the grana are pigment-rich membranes. Partition formation during the development of chloroplasts is correlated with the synthesis of LH (light harvesting) Chl. The synthesis of LH Chl leads to an increase in the size of the photosynthetic unit. In plants, which have to grow in low-light, the limiting factor for photosynthesis is light. Thus, these plants need high pigment concentrations and low enzyme contents. The grana stacks in low-light plants provide the possibility for the co-operation of several antennae systems for the collection of light quanta. This is advantageous, as it allows a more efficient collection of light per electron transport chain. The discrepancy between the absorption power of chlorophyll and the rate capacity of the electron transport system is reduced by the larger size of the photosynthetic unit in low-light plants. An increase in the size of the photosynthetic unit leads to a decrease in the light energy needed to saturate photosynthesis and to a decrease in the photosynthetic rate based on chlorophyll content. Therefore, the differences in the light required for saturation and the light-saturated rate of photosynthesis under high external CO₂-concentration are explained by the variability of the size of the photosynthetic unit. The outer membranes of the grana and the stroma thylakoids contain PS I, NADP⁺-reductase and ATP-synthetase. NADP⁺-reduction and ATP-synthesis only occur at the surface of the o.

L23 ANSWER 89 OF 93 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1978:166887 HCAPLUS

DOCUMENT NUMBER: 88:166887

TITLE: Limited cooperativity in the coupling between electron flow and photosynthetic ATP synthesis. A comparative study in chromatophores phosphorylating at very different rates

AUTHOR(S): Casadio, Rita; Baccarini-Melandri, Assunta; Melandri, Bruno A.

CORPORATE SOURCE: Inst. Bot., Univ. Bologna, Bologna, Italy

SOURCE: FEBS Letters (1978), 87(2), 323-8
CODEN: FEBLAL; ISSN: 0014-5793
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Utilizing highly active chromatophore preps. from Rhodopseudomonas capsulata (strain Kbl), the relations between the rate of ATP synthesis and the extent of Δp were re-examined and the results compared with those obtained in chromatophores from the same strain, in which the ability for ATP synthesis had been drastically impaired either by partial detachment of the coupling factor or by treatment with the covalent inhibitor DCCD(N,N'-dicyclohexylcarbodiimide). Based on the average size of a photosynthetic unit (100 mols. bacteriochlorophyll (BChl)/reaction center) and the turnover time of the cyclic electron transport system (of the order of 10 ms), the rate of photophosphorylation observed in the preps. used in this study (600-900 $\mu\text{mol/h/mg}$ BChl) corresponded to a theor. ATP/2e- value of 0.36-0.55. This indicates that a large percentage of photosynthetic units are associated with an ATP synthetase complex.

L23 ANSWER 90 OF 93 MEDLINE on STN DUPLICATE 25
ACCESSION NUMBER: 76184796 MEDLINE
DOCUMENT NUMBER: PubMed ID: 178369
TITLE: Gramicidin S-synthetase. A further characterization of phenylalanine racemase, the light enzyme of gramicidin s-synthetase.
AUTHOR: Vater J; Kleinkauf H
SOURCE: Biochimica et biophysica acta, (1976 May 13) 429 (3) 1062-72.
Journal code: 0217513. ISSN: 0006-3002.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 197608
ENTRY DATE: Entered STN: 19900313
Last Updated on STN: 19980206
Entered Medline: 19760802

AB 1. Chromatography on hydroxyapatite and on aminoethyl-Sepharose as well as isoelectric focusing were introduced as new effective purification procedures for phenylalanine racemase (EC 5.1.1.11). The enzyme preparations obtained were essentially homogeneous, as demonstrated by specific activity measurements and polyacrylamide gel electrophoresis. 2. The enzyme is not dissociable by sodium dodecyl sulfate. 3. Phenylalanine racemase is an acidic protein with an isoelectric point of approx. 4.6 (isoelectric focusing). 4. The Michaelis constants of L-Phe and D-Phe in the aminoacyl adenylate activation are 0.06 and 0.13 mM, respectively. 5. From our studies with structural analogues of phenylalanine we infer that the amino group of this amino acid is essential for its binding to the aminoacyl adenylate reaction center. The carboxyl group is not at all or only weakly bound. The benzene ring of phenylalanine which determines substrate recognition also seems to be of minor importance for substrate binding.

L23 ANSWER 91 OF 93 HCAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 1973:502015 HCAPLUS
DOCUMENT NUMBER: 79:102015
TITLE: Polypyrrroles formed from porphobilinogen and amines by uroporphyrinogen synthetase of Rhodopseudomonas spheroides
AUTHOR(S): Davies, Richard C.; Neuberger, Albert
CORPORATE SOURCE: Dep. Chem. Pathol., St. Mary's Hosp. Med. Sch., London, UK
SOURCE: Biochemical Journal (1973), 133(3), 471-92
CODEN: BIJOAK; ISSN: 0264-6021
DOCUMENT TYPE: Journal
LANGUAGE: English
AB The effect of NH_2OMe , NH_2OH , and NH_3 on the action of uroporphyrinogen I synthetase from R. spheroides NCIB 8253 was studied. The purified enzyme had a mol. weight of 36,000 and was a single polypeptide chain,

migrating as an active band on disc electrophoresis at pH 7.5 and 8.9. When the amines were present, normal formation of uroporphyrinogen from porphobilinogen at pH 8.2 was replaced by synthesis of open-chain polypyrroles with unsubstituted α -pyrrolic positions. The inhibitory amines were incorporated into the pyrroles by exchange with the NH₂ function of the original porphobilinogen (NH₂OH:NH₂OMe:NH₃ = 10:5:1). Studies on a labeled NH₂OH pyrrole suggested a noncyclized tetrapyrrolic structure, probably a pyrrolylmethane. On nonenzymic cyclization the polypyrroles gave porphyrins and the amines were released. With NH₂OH only, another modified pyrrole, probably a monopyrrole, was detected. Thus, the amines appear to inhibit the cyclization step of the enzyme-catalyzed reaction with the possible involvement of pos. charged reaction centers.

L23 ANSWER 92 OF 93 NTIS COPYRIGHT 2005 NTIS on STN
 ACCESSION NUMBER: 1995(16):04820
 NTIS ORDER NUMBER: DE95002797/XAB
 TITLE: Nitrogen control of chloroplast differentiation.
 AUTHOR: Schmidt, G. W.
 CORPORATE SOURCE: Georgia Univ. Research Foundation, Inc., Athens.
 Sponsor: Department of Energy, Washington, DC.
 (071618000 9511609)
 NUMBER OF REPORT: DE95002797/XAB; DOE/ER/13188-9
 6p; 1994
 NUMBER OF CONTRACT: FG09-84ER13188
 CONTROLLED TERM: Report
 COUNTRY: United States
 LANGUAGE: English
 NOTES: Sponsored by Department of Energy, Washington, DC.
 AVAILABILITY: Order this product from NTIS by: phone at
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 countries); fax at (703)605-6900; and email at
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 NTIS Prices: PC A02/MF A01

OTHER SOURCE: GRA&I9509; ERA9512

AB This project was directed toward understanding how the availability of nitrogen affects the accumulation of chloroplast pigments and proteins that function in energy transduction and carbon metabolism. The availability of this nutrient most pervasively limits plant growth and agricultural productivity but the molecular and physiological consequences of nitrogen-deficiency are poorly understood. The model system for our studies of nitrogen-dependent regulation of chloroplast differentiation is the unicellular green alga *Chlamydomonas reinhardtii* which is grown phototrophically in a continuous culture system. When 150 (μ)M nitrogen is provided at a dilution rate of 0.25 volumes of the growth medium per day, the cultures are sustained at a density of less than 10(^{sup} 5) cells/ml and chlorophyll deficiency, the classical symptom of nitrogen-deficiency, becomes quite pronounced. We found that there is a concomitant loss of light-harvesting complexes and reduced levels of Photosystem II reaction center complexes while ATP synthetase and Photosystem I reaction centers are maintained at high levels. Moreover, reduced rates of chloroplast protein synthesis are due to differential effects on mRNA translation. In contrast, the deficiency of light-harvesting genes is due to marked reductions of the nuclear-encoded cab mRNAs. Although there is no significant reduction of the amounts of RuBPCase, we also detected substantial changes in the mRNA abundance of the alga's two small subunit genes. All of the effects of nitrogen-limitation are readily reversible: greening of cells is completed within 24 hours after provision of 10 mM ammonium. During this time, the plastid translational constraints are disengaged and progressive changes in the abundance of nuclear transcripts occur, including a transient 30-fold elevation of (und cab) mRNAs.

L23 ANSWER 93 OF 93 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN
 ACCESSION NUMBER: 1982:99433 BIOSIS
 DOCUMENT NUMBER: PREV198223029425; BR23:29425

TITLE: PATHWAYS OF GENERATION AND UTILIZATION OF ELECTROCHEMICAL
POTENTIAL OF HYDROGEN IONS IN BIO MEMBRANES.

AUTHOR(S): SKULACHEV V P [Reprint author]

CORPORATE SOURCE: DEP BIOENERGETICS, A N BELOZERSKY LAB MOL BIOL BIOORGANIC
CHEM, MOSCOW STATE UNIV, MOSCOW 117234, USSR

SOURCE: Sov. Sci. Rev., Sect. D: Biol. Rev., pp. P83-156.
SKULACHEV, V. P. (ED.). SOVIET SCIENTIFIC REVIEWS SECTION
D: BIOLOGY REVIEWS, VOL. 1. IX+476P. HARWOOD ACADEMIC
PUBLISHERS: NEW YORK, N.Y., USA; CHUR, SWITZERLAND. ILLUS.
1980 (RECD. 1981).
Publisher: Series: Soviet Scientific Reviews Section D
Biology Reviews.
CODEN: SRSRDL. ISSN: 0143-0424. ISBN: 3-7186-0020-A.

DOCUMENT TYPE: Book

FILE SEGMENT: BR

LANGUAGE: ENGLISH

=> d his

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS,
LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

L1 4 S ASTAXANTHIN (W) SYNTHASE?

L2 0 S ASTAXANTHIN (W) SYNTHETASE?

L3 6416 S ASTAXANTHIN

L4 24046 S "AST"

L5 194127 S SYNTHETASE?

L6 0 S L3 (W) L5

L7 39 S L3 AND L5

L8 24 DUP REM L7 (15 DUPLICATES REMOVED)

L9 41050 S REACTION (A) CENTER

L10 0 S L8 AND L9

L11 153 S L5 AND L9

L12 371127 S REDUCTASE?

L13 21 S L11 AND L12

L14 19 DUP REM L13 (2 DUPLICATES REMOVED)

L15 7593 S P450 REDUCTASE?

L16 0 S L9 AND L15

L17 0 S L4 AND L9

L18 59612 S "BETA-CAROTENE"

L19 64 S L4 AND L18

L20 0 S L9 AND L19

L21 0 S L1 AND L9

L22 3 DUP REM L1 (1 DUPLICATE REMOVED)

L23 93 DUP REM L11 (60 DUPLICATES REMOVED)

=> s l23 60-80 ibib ab

MISSING OPERATOR L23 60-80

The search profile that was entered contains terms or
nested terms that are not separated by a logical operator.

=> d l23 60-80 ibib ab

L23 ANSWER 60 OF 93 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on
STN

ACCESSION NUMBER: 1994:499226 SCISEARCH

THE GENUINE ARTICLE: PB676

TITLE: NITROGEN-METABOLISM IN SENESCING LEAVES

AUTHOR: FELLER U (Reprint); FISCHER A

CORPORATE SOURCE: UNIV BERN, INST PLANT PHYSIOL, ALTENBERGRAIN 21, CH-3013
BERN, SWITZERLAND (Reprint)

COUNTRY OF AUTHOR: SWITZERLAND

SOURCE: CRITICAL REVIEWS IN PLANT SCIENCES, (1994) Vol. 13, No. 3,
pp. 241-273.
ISSN: 0735-2689.

PUBLISHER: CRC PRESS INC, 2000 CORPORATE BLVD NW, JOURNALS CUSTOMER
SERVICE, BOCA RATON, FL 33431.

DOCUMENT TYPE: General Review; Journal
FILE SEGMENT: AGRI
LANGUAGE: English
REFERENCE COUNT: 350
ENTRY DATE: Entered STN: 1994
Last Updated on STN: 1994

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Leaf senescence is a highly organized process and not a passive decay. Photosynthesizing mesophyll cells lose their functions in an early phase, while the epidermal layer with the stomates and the phloem remains functional throughout senescence. The subcellular compartmentation is maintained and allows the cooperation of different organelles in the remobilization of constituents. Nitrogen metabolism changes at the onset of senescence from assimilation to remobilization. Enzymes involved in nitrate reduction are lost, while some enzymes of intermediary nitrogen metabolism are maintained longer, and some catabolic enzymes reach highest activities during senescence. Chloroplasts are dismantled early, but mitochondria remain active and may fuel remobilization processes. Chloroplast proteins are degraded, and this nitrogen fraction can be translocated via the phloem from senescing leaves to sinks within the same plant. In contrast, chlorophyll is degraded, fragments produced reach the vacuole, and catabolites accumulate there. Nuclear DNA is maintained until a very late phase. The export of nitrogen from senescing plant parts is important for the economic use of this macronutrient. The regulation of senescence at the whole plant level as well as at the molecular level is only rudimentarily known, although interesting new aspects have been presented recently.

L23 ANSWER 61 OF 93 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1994:205118 HCAPLUS

DOCUMENT NUMBER: 120:205118

TITLE: Laue image analysis. II. Variable radial elliptical masking and its application in studies involving inherently streaked Laue exposures

AUTHOR(S): Greenhough, Trevor J.; Shrive, Annette K.

CORPORATE SOURCE: Dep. Phys., Univ. Keele, Keele/Staffordshire, ST5 5BG, UK

SOURCE: Journal of Applied Crystallography (1994), 27(1), 111-21

CODEN: JACGAR; ISSN: 0021-8898

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Many Laue diffraction patterns from crystals of particular biol. or chemical interest are of insufficient quality for their anal. to be feasible. In many cases, this is because of pronounced streaking of the spots due to either large mosaic spread or disorder introduced during reactions in the crystal. Methods for the anal. of exposures exhibiting radial or near-radial streaking are described, along with their application in Laue diffraction studies of form-II crystals of Met-tRNA synthetase and a photosynthetic reaction center from Rhodospirillum rubrum sphaeroides. In both cases, variable elliptical radial masking has led to significant improvements in data quality and quantity and exposures that previously were too streaked to process may now be analyzed. These masks can also provide circular profiles as a special case for processing high-quality Laue exposures and spatial-overlap deconvolution may be performed using the elliptical or circular masks.

L23 ANSWER 62 OF 93 MEDLINE on STN

ACCESSION NUMBER: 93252952 MEDLINE

DOCUMENT NUMBER: PubMed ID: 8486714

TITLE: Methyl jasmonate-regulated translation of nuclear-encoded chloroplast proteins in barley (Hordeum vulgare L. cv. salome).

AUTHOR: Reinbothe S; Reinbothe C; Parthier B

CORPORATE SOURCE: Institute of Plant Biochemistry, Halle/Saale, Federal Republic of Germany.

SOURCE: Journal of biological chemistry, (1993 May 15) 268 (14) 10606-11.

Journal code: 2985121R. ISSN: 0021-9258.

PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199306
ENTRY DATE: Entered STN: 19930618
Last Updated on STN: 19980206
Entered Medline: 19930608

AB The naturally occurring plant growth regulator (-)-jasmonic acid methyl ester (JaMe) induces the formation of novel abundant proteins in excised barley leaf segments. Concomitantly, this substance depresses the translation of most preexisting ("control") leaf mRNAs, including those for nuclear-encoded chloroplast proteins such as the small subunit of ribulose-1,5-bisphosphate carboxylase/oxygenase (SSU, rbcS gene product) and several light harvesting chlorophyll protein complex apoproteins (LHCPs, cab gene products). The changes in protein synthesis observed for SSU and LHCPs did not correspond to equivalent alterations in the rbcS and cab transcript levels. Analysis of polysome-associated in vitro translatable and hybridizable mRNAs, however, demonstrated a restriction of rbcS and cab transcripts to smaller polysomes in JaMe-exposed leaf tissues, in comparison to water-treated tissues. Since treatment of JaMe-incubated leaf segments with cycloheximide prior to harvest led to a shift of both transcripts toward larger polysomes, a hormone-induced impairment of chain initiation is assumed to lower translation of SSU and LHCP in situ. In contrast, the mRNA for plastid leucyl-tRNA synthetase (LRS1, lrs1 gene product) neither changed its abundance nor its association with polysomes in JaMe-treated leaves and was translated into the corresponding polypeptide. Together, our results highlight a remarkable variability of nuclear gene expression in response to plant growth regulators of the methyl jasmonate type.

L23 ANSWER 63 OF 93 MEDLINE on STN
ACCESSION NUMBER: 93272335 MEDLINE
DOCUMENT NUMBER: PubMed ID: 8388782
TITLE: Calcium/calmodulin-dependent and -independent phytochrome signal transduction pathways.
COMMENT: Erratum in: Cell 1994 Nov 18;79(4):743
AUTHOR: Neuhaus G; Bowler C; Kern R; Chua N H
CORPORATE SOURCE: Laboratory of Plant Molecular Biology, Rockefeller University, New York, New York 10021-6399.
CONTRACT NUMBER: 44640
SOURCE: Cell, (1993 Jun 4) 73 (5) 937-52.
Journal code: 0413066. ISSN: 0092-8674.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199306
ENTRY DATE: Entered STN: 19930716
Last Updated on STN: 20000303
Entered Medline: 19930630

AB Phytochrome is a well-characterized plant photoreceptor, able to modulate many morphological, physiological, and biochemical events through as yet undefined mechanisms. By developing single-cell assays to visualize phytochrome responses, we have studied the effects of microinjecting putative signaling intermediates into phytochrome-deficient tomato cells. We demonstrate that phytochrome phototransduction initially involves the activation of one or more G proteins that are coupled to at least two different pathways; one pathway requires calcium and activated calmodulin and can stimulate expression of a photoregulated cab-GUS reporter gene together with the synthesis and assembly of some, but not all, of the photosynthetic complexes. The other pathway, controlling anthocyanin biosynthesis, does not require calcium. Furthermore, our results reveal that phytochrome signaling is cell autonomous and is not likely to require any light-activated steps downstream of the G protein.

L23 ANSWER 64 OF 93 HCAPLUS COPYRIGHT 2005 ACS on STN
ACCESSION NUMBER: 1994:187374 HCAPLUS
DOCUMENT NUMBER: 120:187374

TITLE: Chloroplast thylakoid protein changes induced by low growth temperature in maize revealed by immunocytochemistry

AUTHOR(S): Robertson, E. J.; Baker, N. R.; Leech, R. M.

CORPORATE SOURCE: Dep. Biol., Univ. York, Heslington/York, YO1 5DD, UK

SOURCE: Plant, Cell and Environment (1993), 16(7), 809-18
CODEN: PLCEDV; ISSN: 0140-7791

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Tissue-specific effects of low growth temperature on maize chloroplast thylakoid protein accumulation were analyzed using immunocytochemistry. Sections of leaves from plants grown at 25 and 14°C were probed with antibodies to specific chloroplast thylakoid proteins from the four major protein multisubunit complexes of the thylakoid membrane followed by fluorescein-conjugated goat antirabbit antibodies. At a normal growth temperature of 25°C, the 32 kDa D1 protein of the photosystem II reaction center and the 33 kDa protein of the extrinsic oxygen-evolving complex of photosystem II are both accumulated to a greater degree in the mesophyll than in the bundle sheath chloroplasts. In contrast, subunit II of photosystem I, cytochrome f and the α - and β -subunits of ATP synthetase are predominant in the bundle sheath thylakoids at 25°C. A striking difference between the 25°C-grown and the 14°C-grown leaf tissue was the presence in the latter of (20-30%) cells whose chloroplasts apparently completely lack several of the thylakoid proteins. In plants grown at 14°C, the accumulation of the 33 kDa protein of the extrinsic oxygen-evolving complex of photosystem II was apparently unchanged, but other thylakoid proteins showed a significant reduction. The uneven distribution of proteins between the bundle sheath and mesophyll chloroplasts observed at 25°C was also maintained at 14°C. Reduction in the fluorescence at 14°C was manifested either as an overall reduction in the diffuse fluorescence across the chloroplast profiles or less frequently as a reduction to small discrete bodies of intense fluorescence. The significance of these results to low-temperature-induced reduction in the photosynthetic productivity of maize is discussed.

L23 ANSWER 65 OF 93 MEDLINE on STN DUPLICATE 14

ACCESSION NUMBER: 93307499 MEDLINE

DOCUMENT NUMBER: PubMed ID: 8319807

TITLE: Analysis of a mutant amino acid-activating domain of surfactin synthetase bearing a serine-to-alanine substitution at the site of carboxylthioester formation.

AUTHOR: Vollenbroich D; Kluge B; D'Souza C; Zuber P; Vater J

CORPORATE SOURCE: Institut für Biochemie und Molekulare Biologie, Technische Universität Berlin, Germany.

SOURCE: FEBS letters, (1993 Jul 5) 325 (3) 220-4.
Journal code: 0155157. ISSN: 0014-5793.

PUB. COUNTRY: Netherlands

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199308

ENTRY DATE: Entered STN: 19930813
Last Updated on STN: 19980206
Entered Medline: 19930805

AB The reactive serine of the TGGHSL thioester binding motif of the first amino acid-activating domain of surfactin synthetase was replaced by alanine using site-directed mutagenesis. The multienzyme from cells of the resulting mutant lost its ability for thioester formation with L-Glu and was therefore inactive in surfactin production. The thiolation reactions catalyzed by the other amino acid-activating domains of surfactin synthetase were not affected by the mutation. The results show that L-Glu is activated at the first domain of surfactin synthetase, and give further evidence that a serine residue is essential for substrate amino acid activation at the reaction centers of peptide synthetases.

L23 ANSWER 66 OF 93 MEDLINE on STN DUPLICATE 15

ACCESSION NUMBER: 92078181 MEDLINE

DOCUMENT NUMBER: PubMed ID: 1744112

TITLE: An active serine is involved in covalent substrate amino acid binding at each reaction center of gramicidin S synthetase.

AUTHOR: Schlumbohm W; Stein T; Ullrich C; Vater J; Krause M; Marahiel M A; Kruff V; Wittmann-Liebold B

CORPORATE SOURCE: Institute of Biochemistry and Molecular Biology, Technical University of Berlin, Federal Republic of Germany.

SOURCE: Journal of biological chemistry, (1991 Dec 5) 266 (34) 23135-41.

Journal code: 2985121R. ISSN: 0021-9258.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199201

ENTRY DATE: Entered STN: 19920202
Last Updated on STN: 19980206
Entered Medline: 19920113

AB The condensing peptide forming multienzyme of gramicidin S synthetase (gramicidin S synthetase 2) was specifically labeled at its putative thiotemplate sites for L-valine and L-leucine by covalent incorporation of the ¹⁴C-labeled substrate amino acids. The thioester complexes of the multienzyme were digested with CNBr, Staphylococcus aureus V8 protease, and pepsin. Reaction center peptides containing the [¹⁴C]valine and [¹⁴C]leucine labels were isolated in pure form. They show a high degree of sequence similarity and contain the same consensus sequence LGGH/DXL. The labels were eliminated in the first Edman degradation step. A dehydroalanine was identified which can originate from either a cysteine or a serine. The comparison of the chemical results with the deduced amino acid sequence of the grsB gene encoding the gramicidin S synthetase 2 revealed that 4 such motifs are located within the gene structure, each of them being localized in the 3'-terminal region of one of 4 gene segments grsB1-B4. They have a size of approximately 2 kilobases and presumably code for the 4 amino acid activating domains of the synthetase. Surprisingly a serine was found at each putative substrate amino acid-binding position instead of a cysteine as postulated by the thiotemplate mechanism. Therefore the data suggest that active serine residues are involved in nonribosomal peptide syntheses of microbial peptides.

L23 ANSWER 67 OF 93 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on STN DUPLICATE 16

ACCESSION NUMBER: 1992:9786 BIOSIS

DOCUMENT NUMBER: PREV199293009786; BA93:9786

TITLE: REGULATION OF CHLOROPLAST METABOLISM IN LEAVES EVIDENCE THAT NADP-DEPENDENT GLYCERALDEHYDEPHOSPHATE DEHYDROGENASE BUT NOT FERREDOXIN-NADP REDUCTASE CONTROLS ELECTRON FLOW TO PHOSPHOGLYCERATE IN THE DARK-LIGHT TRANSITION.

AUTHOR(S): SIEBKE K [Reprint author]; LAISK A; NEIMANIS S; HEBER U

CORPORATE SOURCE: INSTITUTE BOTANY PHARMACEUTICAL BIOLOGY, UNIVERSITY WUERZBURG, W-8700 WUERZBURG, W GER

SOURCE: Planta (Heidelberg), (1991) Vol. 185, No. 3, pp. 337-343. CODEN: PLANAB. ISSN: 0032-0935.

DOCUMENT TYPE: Article

FILE SEGMENT: BA

LANGUAGE: ENGLISH

ENTRY DATE: Entered STN: 10 Dec 1991
Last Updated on STN: 6 Mar 1992

AB P700 is rapidly, but only transiently photooxidized upon illuminating dark-adapted leaves. Initial oxidation is followed by a reductive phase even under far-red illumination which excites predominantly photosystem (PS) I. In this phase, oxidized P700 is reduced by electrons coming from PSII. Charge separation in the reaction center of PSI is prevented by the unavailability of electron acceptors on the reducing side of PSI. It is subsequently made possible by the opening of an electron gate which is situated between PSI and the electron acceptor phosphoglycerate. Electron acceptors immediately available for reduction while the gate is closed corresponded to 10 nmol · (mg

chlorophyll)-1 electrons in geranium leaves, 16 nmol · (mg chlorophyll)⁻¹ in sunflower and 22 nmol · (mg chlorophyll)⁻¹ in oleander. Reduction of NADP during the initial phase of P700 oxidation showed that the electron gate was not represented by ferredoxin-NADP reductase. Availability of ATP indicated that electron flow was not hindered by deactivation of the thylakoid ATP synthetase. It is concluded that NADP-dependent glyceraldehydephosphate dehydrogenase is completely deactivated in the dark and activated in the light. The rate of activation depends on the length of the preceding dark period. As chloroplasts contain both NAD- and NADP-dependent glyceraldehydephosphate dehydrogenases, deactivation of the NADP-dependent enzyme disconnects chloroplast NAD and NADP systems and prevents phosphoglycerate reduction in the dark at the expense of NADPH and ATP which are generated by glucose-6-phosphate oxidation and glycolytic starch breakdown, respectively.

L23 ANSWER 68 OF 93 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1992:16605 HCAPLUS

DOCUMENT NUMBER: 116:16605

TITLE: Promoter recognition by the RNA polymerase from vegetative cells of the cyanobacterium *Anabaena* 7120

AUTHOR(S): Schneider, George J.; Lang, Jean D.; Haselkorn, Robert

CORPORATE SOURCE: Dep. Biochem. Mol. Biol. Mol. Genet. Cell Biol., Univ. Chicago, Chicago, IL, 60637, USA

SOURCE: Gene (1991), 105(1), 51-60
CODEN: GENED6; ISSN: 0378-1119

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The transcription start points (tsp) of 7 genes of *Anabaena* 7120 were previously identified by S1 nuclease protection and primer extension expts. using RNA extracted from cells. In the present work, these tsp were confirmed, with 1 exception, by in vitro transcription using purified. RNA polymerases of *Anabaena* 7120 and *Escherichia coli*, and crude exts. of *Anabaena* 7120 active in transcription. In all cases, the template for transcription consisted of closed circular plasmid DNA in which the putative promoter-containing fragment was cloned in front of a strong terminator, which resulted in defined pseudo-runoff transcripts whose sizes correspond (with one exception) to those expected on the basis of the tsp determined for in vivo RNA. These results, together with other obtained with templates containing bacteriophage T4 or cyanophage N1 promoters, led to the conclusion that the principal *Anabaena* 7120 RNA polymerase prefers promoters whose sequence and spacing approx. that of the *E. coli* consensus promoter, and that the *Anabaena* 7120 genes expressed in vegetative cells, characterized to date, have relatively weak promoters.

L23 ANSWER 69 OF 93 BIOTECHDS COPYRIGHT 2005 THE THOMSON CORP. on STN

ACCESSION NUMBER: 1990-07873 BIOTECHDS

TITLE: Flavo pyruvate-decarboxylase: a semisynthetic enzyme model for pyruvate-oxidase and acetolactate-synthetase; potential application in biocatalyst design

AUTHOR: Annan N; Jordan F

LOCATION: Department of Chemistry, Rutgers, The State University of New Jersey, Newark, New Jersey 07102, USA.

SOURCE: J.Am.Chem.Soc.; (1990) 112, 8, 3222-23
CODEN: JACSAT

DOCUMENT TYPE: Journal

LANGUAGE: English

AB In order to gain information on biocatalyst design by alteration of the catalytic environment of an enzyme to change its reaction specificity, a flavin analog was successfully incorporated into the active site of pyruvate-decarboxylase (EC-4.1.1.1). The modified enzyme was competent in redox reactions and performed the reactions anticipated for pyruvate-oxidase (EC-1.2.3.3). 4 10-methylisoalloxazine analogs were synthesized. This is the first demonstration of the diversion of the fate of a common intermediate by covalent incorporation of an oxidative cofactor. At a concentration of 1 mM, 8- α -(bromoacetyl)-10-methylisoalloxazine totally and irreversibly inactivated pure pyruvate-decarboxylase with respect to acetaldehyde formation at pH 6

with a half-life of 2 min. Inactivation was probably due to inactivation of the Cys near to the reaction center. The 6-alpha-bromoacetyl analog was less effective (half-life 4 min, but only 80% inactivation resulted), while the 6-acetyl and 8-acetyl derivatives gave no inhibition. Pyruvate-decarboxylase may be a good model for acetolactate-synthetase and pyruvate-oxidase. (31 ref)

L23 ANSWER 70 OF 93 MEDLINE on STN DUPLICATE 17
ACCESSION NUMBER: 91017465 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2217141
TITLE: The interpretation of site-directed mutagenesis experiments by linear free energy relations.
AUTHOR: Straub J E; Karplus M
CORPORATE SOURCE: Department of Chemistry, Harvard University, Cambridge, MA 02138.
SOURCE: Protein engineering, (1990 Aug) 3 (8) 673-5.
Journal code: 8801484. ISSN: 0269-2139.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199011
ENTRY DATE: Entered STN: 19910117
Last Updated on STN: 19980206
Entered Medline: 19901115

AB Fersht and co-workers have applied a linear free energy relation (Bronsted equation) to analyze site-directed mutagenesis experiments involving the enzyme tyrosyl-tRNA synthetase and have suggested that the Bronsted exponent is linearly correlated with the value of the reaction coordinate at the transition state. We point out that when the mutants differ solely through the formation or deletion of a hydrogen bond away from the reaction center, a linear free energy relation is expected only in limiting cases for which the Bronsted relation exponent is 0, 1 or infinity. The results may be correlated with a conformational coordinate but not with the development of the reaction coordinate per se.

L23 ANSWER 71 OF 93 MEDLINE on STN
ACCESSION NUMBER: 90249505 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2110911
TITLE: Modification of a glnB-like gene product by photosynthetic electron transport in the cyanobacterium Synechococcus 6301.
AUTHOR: Harrison M A; Keen J N; Findlay J B; Allen J F
CORPORATE SOURCE: Department of Pure and Applied Biology, University of Leeds, UK.
SOURCE: FEBS letters, (1990 May 7) 264 (1) 25-8.
Journal code: 0155157. ISSN: 0014-5793.
PUB. COUNTRY: Netherlands
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199006
ENTRY DATE: Entered STN: 19900720
Last Updated on STN: 19950206
Entered Medline: 19900620

AB Covalent modification of a 13 kDa soluble-phase protein occurs during adaptation of cells of the cyanobacterium Synechococcus 6301 (mutant AN112) to light specifically absorbed by photosystem II. This adaptation is accompanied by functional changes indicative of altered excitation energy distribution between the photosystems. The 13 kDa protein is identified by solid-phase N-terminal sequencing as a protein related to PII, the glnB gene product of E. coli. In E. coli, the PII protein undergoes uridylylation and acts as a regulator of glutamine synthetase at both the post-translational and transcriptional levels. The implications of modification of a transcriptional regulator by photosynthetic electron transport are discussed.

L23 ANSWER 72 OF 93 MEDLINE on STN

ACCESSION NUMBER: 88312649 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3137030
TITLE: A mathematical model of the Calvin photosynthesis cycle.
AUTHOR: Pettersson G; Ryde-Pettersson U
CORPORATE SOURCE: Avdelningen for Biokemi, Lunds Universitet, Sweden.
SOURCE: European journal of biochemistry / FEBS, (1988 Aug 15) 175
(3) 661-72.
Journal code: 0107600. ISSN: 0014-2956.
PUB. COUNTRY: GERMANY, WEST: Germany, Federal Republic of
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198810
ENTRY DATE: Entered STN: 19900308
Last Updated on STN: 19970203
Entered Medline: 19881011

AB 1. A mathematical model is presented for photosynthetic carbohydrate formation in C3 plants under conditions of light and carbon dioxide saturation. The model considers reactions of the Calvin cycle with triose phosphate export and starch production as main output processes, and treats concentrations of NADPH, NAD⁺, CO₂, and H⁺ as fixed parameters of the system. Using equilibrium approximations for all reaction steps close to equilibrium steady-state and transient-state relationships are derived which may be used for calculation of reaction fluxes and concentrations of the 13 carbohydrate cycle intermediates, glucose 6-phosphate, glucose 1-phosphate, ATP, ADP, and inorganic (ortho)phosphate. 2. Predictions of the model were examined with the assumption that photosynthate export from the chloroplast occurs to a medium containing orthophosphate as the only exchangeable metabolite. The results indicate that the Calvin cycle may operate in a single dynamically stable steady state when the external concentration of orthophosphate does not exceed 1.9 mM. At higher concentrations of the external metabolite, the reaction system exhibits overload breakdown; the excessive rate of photosynthate export deprives the system of cycle intermediates such that the cycle activity progressively approaches zero. 3. Reactant concentrations calculated for the stable steady state that may obtain are in satisfactory agreement with those observed experimentally, and the model accounts with surprising accuracy for experimentally observed effects of external orthophosphate on the steady-state cycle activity and rate of starch production. 4. Control analyses are reported which show that most of the non-equilibrium enzymes in the system have a strong regulatory influence on the steady-state level of all of the cycle intermediates. Substrate concentration control coefficients for cycle enzymes may be positive, such that an increase in activity of an enzyme may raise the steady-state concentration of the substrate it consumes. 5. Under optimal external conditions (0.15-0.5 mM orthophosphate), reaction flux in the Calvin cycle is controlled mainly by ATP synthetase and sedoheptulose biphosphatase; the cycle activity approaches the maximum velocity that can be supported by the latter enzyme. At lower concentrations of external orthophosphate the cycle activity is controlled almost exclusively by the phosphate translocator. (ABSTRACT TRUNCATED AT 400 WORDS)

L23 ANSWER 73 OF 93 MEDLINE on STN
ACCESSION NUMBER: 89049169 MEDLINE
DOCUMENT NUMBER: PubMed ID: 3056272
TITLE: Control of bacteriochlorophyll accumulation by light in an aerobic photosynthetic bacterium, Erythrobacter sp. OCh 114.
AUTHOR: Shioi Y; Doi M
CORPORATE SOURCE: Division of Biology, Miyazaki Medical College, Japan.
SOURCE: Archives of biochemistry and biophysics, (1988 Nov 1) 266
(2) 470-7.
Journal code: 0372430. ISSN: 0003-9861.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198811
ENTRY DATE: Entered STN: 19900308

Last Updated on STN: 19980206

Entered Medline: 19881130

AB The effect of light on bacteriochlorophyll (Bchl) accumulation as well as the activity of two enzymes in the initial step of the tetrapyrrole biosynthetic pathway was examined in an aerobic photosynthetic bacterium, *Erythrobacter* sp. strain OCh 114. Light clearly regulated the Bchl and carotenoid accumulation, completely suppressing their levels at high light intensity. However, porphyrin and Bchl precursors were not found in either the cells or the growth medium of lighted culture. The level of Bchl showed an inverse relationship to the light energy flux. Kinetic studies showed a Hill coefficient of $n = 3.3$ ($r = 0.973$), indicating a positive cooperativity. Bchl accumulation was stopped immediately upon illumination without any lag or overshoot. Despite low Bchl content, the activities of 5-aminolevulinic acid synthetase and porphobilinogen synthase were rather stimulated, but not suppressed by light. The high activity of enzymes coincided with the results that heme contents, particularly cytochrome c and catalase activity, were increased in light-grown cells. These results suggest that light regulated Bchl accumulation, but not Bchl biosynthesis and that the effect of light is to render newly formed pigment molecules unstable.

L23 ANSWER 74 OF 93 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on
STN DUPLICATE 18

ACCESSION NUMBER: 1988:335624 BIOSIS

DOCUMENT NUMBER: PREV198886042175; BA86:42175

TITLE: THYLAKOID MEMBRANE ELECTROCHEMICAL MECHANISMS OF
PHOTOSYNTHESIS THE MECHANISM OF OXYGEN EVOLUTION IN THE
REACTION CENTER OF PHOTOSYSTEM II OF
GREEN PLANTS.

AUTHOR(S): VOLKOV A G [Reprint author]

CORPORATE SOURCE: AN FRUMKIN INST ELECTROCHEM, ACAD SCI USSR, MOSCOW, USSR

SOURCE: Biologicheskije Membrany (Moscow), (1987) Vol. 4, No. 9, pp.
984-993.

CODEN: BIMEE9. ISSN: 0233-4755.

DOCUMENT TYPE: Article

FILE SEGMENT: BA

LANGUAGE: RUSSIAN

ENTRY DATE: Entered STN: 21 Jul 1988

Last Updated on STN: 21 Jul 1988

AB The four-electron and two-electron mechanisms of water photooxidation in the reaction centers of photosystem II are proposed.

It is supposed that water can be oxidized by the oxygen-evolving complex consisting of four chlorophyll molecules, two pheophytin molecules, several plastoquinone molecules and seven polypeptides. The catalytic complex responsible for the oxidation of water includes four manganese ions. The proton partition between the inner and outer sides of the thylakoid membrane is controlled by the pool of plastoquinones, lipids, polar groups of proteins and ATP-synthetase.

L23 ANSWER 75 OF 93 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on
STN DUPLICATE 19

ACCESSION NUMBER: 1987:299262 BIOSIS

DOCUMENT NUMBER: PREV198784029294; BA84:29294

TITLE: PHOTOSYNTHETIC RESPONSES OF PISUM-SATIVUM TO AN INCREASE IN
IRRADIANCE DURING GROWTH II. THYLAKOID MEMBRANE COMPONENTS.

AUTHOR(S): CHOW W S [Reprint author]; ANDERSON J M

CORPORATE SOURCE: DIV PLANT INDUSTRY, CSIRO, GPO BOX 1600, CANBERRA, ACT
2601, AUSTRALIA

SOURCE: Australian Journal of Plant Physiology, (1987) Vol. 14, No.
1, pp. 9-20.

CODEN: AJPPCH. ISSN: 0310-7841.

DOCUMENT TYPE: Article

FILE SEGMENT: BA

LANGUAGE: ENGLISH

ENTRY DATE: Entered STN: 6 Jul 1987

Last Updated on STN: 6 Jul 1987

AB Following the transfer of pea plants grown at low irradiance (60 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$, 16 h light/8 h dark cycles) to high irradiance (390 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$), the extents and time courses of the increase in

the concentrations of thylakoid membrane components on a chlorophyll basis have been determined. The increase in cytochrome f (.apprx. 70%) and plastoquinone (.apprx. 50%) contents occurred with no noticeable lag phase. The increase in photosystem II reaction centers (PS II, .apprx. 35%) and ATP synthetase (.apprx. 90%) occurred possibly with a lag period of 1-2 days. In contrast, there was no significant increase in the concentration of P700 (reaction center) of PS I complex. The concentration of PS II reaction centers measured by atrazine-binding exceeded that from the O₂ yield per single-turnover flash by a factor of 1.17 (compared with the expected value of 1.14); this contrasts with the factor of 1.8 obtained by P. A. Jursinic and R. Dennenberg [Arch. Biochem. Biophys. (1985) 241, 540-9]. It is suggested that both methods are equivalent for the determination of PS II reaction centers in active chloroplasts. The stoichiometry of PS II:cyt f:PS I was highly flexible, and not fixed at 1:1:1. We obtained the stoichiometries of 1.25:0.7:1.0 for low-light pea chloroplasts and 1.7:1.25:1.0 for chloroplasts in pea plants that had been transferred to high light for about 10 days, demonstrating the dynamic nature of thylakoid composition and function. In the first 2 days after transferring low light pea plants to high light, the time course of the increase in CO₂- and light-saturated rate of leaf photosynthesis corresponded better with that of cyt f and plastoquinone than that of other chloroplast components examined. This suggests that, during the transition period, the relatively prompt increase of cyt b/f and plastoquinone plays a part in enhancing the CO₂- and light-saturated rate of leaf photosynthesis.

L23 ANSWER 76 OF 93 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1988:90010 HCAPLUS

DOCUMENT NUMBER: 108:90010

TITLE: The mechanism of cytochrome oxidase and other reaction centers for electron/proton pumping

AUTHOR(S): Williams, R. J. P.

CORPORATE SOURCE: Inorg. Chem. Lab., Univ. Oxford, Oxford, OX1 3QR, UK

SOURCE: FEBS Letters (1987), 226(1), 1-7

CODEN: FEBLAL; ISSN: 0014-5793

DOCUMENT TYPE: Journal; General Review

LANGUAGE: English

AB The functional significance of the metal centers of cytochrome oxidase is deduced from the ways in which the centers are bound into its peptides. To this end, use is made of structural knowledge of other metalloproteins for O₂ binding (hemocyanin and Hb) and for electron transfer (cytochrome b and azurin). The order and manner in which the motions of helical sections of the oxidase are linked to proton pumping are suggested and a comparison is made with other proton pumps, e.g., ATP synthetases. Exptl. data are reviewed.

L23 ANSWER 77 OF 93 HCAPLUS COPYRIGHT 2005 ACS on STN

ACCESSION NUMBER: 1986:530862 HCAPLUS

DOCUMENT NUMBER: 105:130862

TITLE: The electrochemical mechanism of photosystem II functioning in chloroplasts

AUTHOR(S): Volkov, G.

CORPORATE SOURCE: A. N. Frumkin Inst. Electrochem., Moscow, 117071, USSR

SOURCE: Journal of Electroanalytical Chemistry and Interfacial Electrochemistry (1986), 205(1-2), 245-57

CODEN: JEIEBC; ISSN: 0022-0728

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The electrochem. aspects of O₂ evolution in photosynthesis are considered. A 4-electron mechanism is proposed for the photooxidn. of water in the reaction center of photosystem II of chloroplasts. It is supposed that water can be oxidized by a reaction center consisting of 4 chlorophyll mols., a Mn cluster, 2 pheophytin mols., and several plastoquinone mols. The catalytic complex responsible for the oxidation of water is assumed to include 4 Mn ions. Two cations are necessary for the catalytic formation of the hydrated

chlorophyll dimer; these can be substituted by other cations of multivalent metals. The partition of protons between the inner and outer side of thylakoid membranes can be controlled by the pool of plastoquinones, lipids, polar groups of proteins, and ATP synthetase. The probability of the 4-electron reaction is analyzed thermodynamically and kinetically. The role of Mn in photosynthesis is discussed.

L23 ANSWER 78 OF 93 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation on
STN DUPLICATE 20

ACCESSION NUMBER: 1986:323741 BIOSIS
DOCUMENT NUMBER: PREV198682048046; BA82:48046
TITLE: ACCUMULATION OF CHLOROPHYLL CHLOROPLASTIC PROTEINS AND
THYLAKOID MEMBRANES DURING REVERSION OF CHROMOPLASTS TO
CHLOROPLASTS IN CITRUS-SINENSIS EPICARP.
AUTHOR(S): MAYFIELD S P [Reprint author]; HUFF A
CORPORATE SOURCE: DEP PLANT SCI, UNIV ARIZONA, TUCSON, ARIZ 85721, USA
SOURCE: Plant Physiology (Rockville), (1986) Vol. 81, No. 1, pp.
30-35.
CODEN: PLPHAY. ISSN: 0032-0889.
DOCUMENT TYPE: Article
FILE SEGMENT: BA
LANGUAGE: ENGLISH
ENTRY DATE: Entered STN: 8 Aug 1986
Last Updated on STN: 8 Aug 1986

AB In vitro culture of pericarp segments from fruit of Citrus sinensis (L.) Osbeck cv Valencia was used to determine the temporal sequence in development of chloroplasts from chromoplasts during regreening of the epicarp. Regreening of chromoplasts closely resembled greening of etioplasts, except that regreening proceeded much more slowly. Chlorophyll, the light-harvesting chlorophyll a/b binding protein of photosystem II, the chlorophyll a/b binding protein of photosystem II, the chlorophyll a binding protein of reaction center P-700 of photosystem I, thylakoid membranes, and adenosine triphosphate synthetase were all detected at very low levels in degreened epicarp. All of these increased in parallel during regreening of the epicarp. Ribulose 1,5-bisphosphate carboxylase (RuBPCase) levels were high in degreened epicarp and declined for the first 10 days of culture before reaccumulating in the regreening segments. Light was necessary for the accumulation of all of the chloroplastic components. A lack of exogenous nitrogen did not prevent the accumulation of any chloroplastic component except RuBPCase, although accumulation of the other components was reduced. Sucrose at 150 millimolar in media lacking nitrogen markedly inhibited the accumulation of chlorophyll and light-harvesting chlorophyll a/b protein.

L23 ANSWER 79 OF 93 MEDLINE on STN
ACCESSION NUMBER: 86164260 MEDLINE
DOCUMENT NUMBER: PubMed ID: 2869943
TITLE: Differences in mRNA levels in Anabaena living freely or in
symbiotic association with Azolla.
AUTHOR: Nierzwicki-Bauer S A; Haselkorn R
CONTRACT NUMBER: GM 21823 (NIGMS)
SOURCE: EMBO journal, (1986 Jan) 5 (1) 29-35.
Journal code: 8208664. ISSN: 0261-4189.
PUB. COUNTRY: ENGLAND: United Kingdom
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 198605
ENTRY DATE: Entered STN: 19900321
Last Updated on STN: 19980206
Entered Medline: 19860514

AB Azolla is a small water fern in whose leaf cavities the filamentous nitrogen-fixing cyanobacterium Anabaena azollae is symbiotically associated. Using cloned genes from Anabaena 7120 for glutamine synthetase (GS), ribulose-1,5-bisphosphate (RuBP) carboxylase, nitrogenase and the 32-kd protein of photosystem II, mRNA levels of the corresponding genes in the Anabaena endosymbiont were studied by Northern

hybridization. In RNA isolated from the endosymbiont there is a 10-fold reduction of GS transcript levels, a greater than 5-fold increase in 32-kd transcript levels and a greater than 5-fold decrease in RuBP carboxylase transcript levels, compared with levels in the free-living *Anabaena azollae*. In the endosymbiont and in heterocysts of the free-living *Anabaena azollae* the *nif H*, *nif D*, and *nif K* genes are transcribed from a single *nif* HDK operon.

L23 ANSWER 80 OF 93 MEDLINE on STN
 ACCESSION NUMBER: 86165318 MEDLINE
 DOCUMENT NUMBER: PubMed ID: 3007236
 TITLE: Chloroplast genes for photosynthetic membrane components of higher plants.
 AUTHOR: Gray J C; Bird C R; Courtice G R; Hird S M; Howe C J; Huttly A K; Phillips A L; Smith A G; Willey D L; Bowman C M; +
 SOURCE: Biochemical Society transactions, (1986 Feb) 14 (1) 25-7. Journal code: 7506897. ISSN: 0300-5127.
 PUB. COUNTRY: ENGLAND: United Kingdom
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 198605
 ENTRY DATE: Entered STN: 19900321
 Last Updated on STN: 19900321
 Entered Medline: 19860520

=> d his

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

L1 4 S ASTAXANTHIN (W)SYNTHASE?
 L2 0 S ASTAXANTHIN (W)SYNTHETASE?
 L3 6416 S ASTAXANTHIN
 L4 24046 S "AST"
 L5 194127 S SYNTHETASE?
 L6 0 S L3 (W)L5
 L7 39 S L3 AND L5
 L8 24 DUP REM L7 (15 DUPLICATES REMOVED)
 L9 41050 S REACTION (A) CENTER
 L10 0 S L8 AND L9
 L11 153 S L5 AND L9
 L12 371127 S REDUCTASE?
 L13 21 S L11 AND L12
 L14 19 DUP REM L13 (2 DUPLICATES REMOVED)
 L15 7593 S P450 REDUCTASE?
 L16 0 S L9 AND L15
 L17 0 S L4 AND L9
 L18 59612 S "BETA-CAROTENE"
 L19 64 S L4 AND L18
 L20 0 S L9 AND L19
 L21 0 S L1 AND L9
 L22 3 DUP REM L1 (1 DUPLICATE REMOVED)
 L23 93 DUP REM L11 (60 DUPLICATES REMOVED)

=> e hoshino t/au

E1 1 HOSHINO SYUUJI/AU
 E2 1 HOSHINO SYUZO/AU
 E3 4600 --> HOSHINO T/AU
 E4 5 HOSHINO T */AU
 E5 1 HOSHINO T K/AU
 E6 1 HOSHINO TADA/AU
 E7 1 HOSHINO TADAFUSA/AU
 E8 21 HOSHINO TADAHIDE/AU
 E9 27 HOSHINO TADAHIKO/AU
 E10 1 HOSHINO TADAMATSU/AU

E11 80 HOSHINO TADAO/AU
E12 60 HOSHINO TADASHI/AU

=> s e3

L24 4600 "HOSHINO T"/AU

=> e kazuyuki o/au

E1 1 KAZUYUKI NAGATA/AU
E2 1 KAZUYUKI NAKAGAWA/AU
E3 6 --> KAZUYUKI O/AU
E4 1 KAZUYUKI OGURA/AU
E5 1 KAZUYUKI OHE/AU
E6 1 KAZUYUKI OHMOTO/AU
E7 1 KAZUYUKI OOISHI/AU
E8 6 KAZUYUKI S/AU
E9 1 KAZUYUKI SAKOTA/AU
E10 1 KAZUYUKI SASAKI/AU
E11 1 KAZUYUKI SHIBATA/AU
E12 1 KAZUYUKI SHIMIZU/AU

=>

=> s e3

L25 6 "KAZUYUKI O"/AU

=> e yutaka s/au

E1 1 YUTAKA R/AU
E2 1 YUTAKA RUKUDA/AU
E3 12 --> YUTAKA S/AU
E4 1 YUTAKA SAITO/AU
E5 1 YUTAKA SANAI/AU
E6 2 YUTAKA SASAKI/AU
E7 1 YUTAKA SAWAMUKAI/AU
E8 2 YUTAKA SAWAYAMA/AU
E9 3 YUTAKA SEIGO/AU
E10 3 YUTAKA SEINO/AU
E11 1 YUTAKA SHIGEHIO/AU
E12 1 YUTAKA SHIOMI/AU

=> s e3

L26 12 "YUTAKA S"/AU

=> s l24 or l25 or l26

L27 4617 L24 OR L25 OR L26

=> d his

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

L1 4 S ASTAXANTHIN (W) SYNTHASE?
L2 0 S ASTAXANTHIN (W) SYNTHETASE?
L3 6416 S ASTAXANTHIN
L4 24046 S "AST"
L5 194127 S SYNTHETASE?
L6 0 S L3 (W) L5
L7 39 S L3 AND L5
L8 24 DUP REM L7 (15 DUPLICATES REMOVED)
L9 41050 S REACTION (A) CENTER
L10 0 S L8 AND L9
L11 153 S L5 AND L9
L12 371127 S REDUCTASE?
L13 21 S L11 AND L12
L14 19 DUP REM L13 (2 DUPLICATES REMOVED)
L15 7593 S P450 REDUCTASE?
L16 0 S L9 AND L15
L17 0 S L4 AND L9

L18 59612 S "BETA-CAROTENE"
 L19 64 S L4 AND L18
 L20 0 S L9 AND L19
 L21 0 S L1 AND L9
 L22 3 DUP REM L1 (1 DUPLICATE REMOVED)
 L23 93 DUP REM L11 (60 DUPLICATES REMOVED)
 E HOSHINO T/AU
 L24 4600 S E3
 E KAZUYUKI O/AU
 L25 6 S E3
 E YUTAKA S/AU
 L26 12 S E3
 L27 4617 S L24 OR L25 OR L26

=> s 15 and 127

L28 19 L5 AND L27

=> dup rem 128

PROCESSING COMPLETED FOR L28

L29 13 DUP REM L28 (6 DUPLICATES REMOVED)

=> d 1-13 ibib ab

L29 ANSWER 1 OF 13 EMBASE COPYRIGHT (c) 2005 Elsevier B.V. All rights
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ACCESSION NUMBER: 2005233116 EMBASE
 TITLE: Isolation of a low-molecular-weight, multicopy plasmid,
 pNHK101, from *Thermus* sp. TK10 and its use as an expression
 vector for *T. thermophilus* HB27.
 AUTHOR: Kobayashi H.; Kuwae A.; Maseda H.; Nakamura A.;
 Hoshino T.
 CORPORATE SOURCE: A. Nakamura, Division of Integrative Environmental
 Sciences, Graduate School of Life and Environmental
 Sciences, University of Tsukuba, Tennodai 1-1-1, Tsukuba,
 Ibaraki 305-8572, Japan. a-nak@agbi.tsukuba.ac.jp
 SOURCE: Plasmid, (2005) Vol. 54, No. 1, pp. 70-79.
 Refs: 27
 ISSN: 0147-619X CODEN: PLSMDX
 PUBLISHER IDENT.: S 0147-619X(04)00145-3
 COUNTRY: United States
 DOCUMENT TYPE: Journal; Article
 FILE SEGMENT: 004 Microbiology
 LANGUAGE: English
 SUMMARY LANGUAGE: English
 ENTRY DATE: Entered STN: 20050616
 Last Updated on STN: 20050616

AB We isolated a small multicopy cryptic plasmid, pNHK101, from *Thermus* sp.
 TK10 for use as a replicon of a *Thermus* expression vector. The nucleotide
 sequence of pNHK101 revealed that this plasmid was 1564 bp long, with a
 total G + C content of 66.8%, which was in agreement with that of *Thermus*
 genomic DNA. The sequence did not show any significant similarities to
 any other plasmids; also, the amino acid sequences of four putative open
 reading frames, found in the plasmid, did not show strong similarities to
 those in the databases, except the ORF1, which had very slight
 similarities to several replication proteins of plasmids from other
 bacteria. pNHK101 was able to replicate in *Thermus thermophilus* HB27 with
 copy number about 80, and was stably maintained at 60°C, but became
 unstable at 70°C. Based on pNHK101, we constructed a plasmid
 vector, pKMH052, containing the highly thermostable kanamycin resistance
 gene as a selective marker. The copy number of pKMH052 decreased to about
 one-fourth of that of pNHK101, but stability at 60°C did not alter
 under non-selective conditions. pKMH052 was compatible with pTT8, and
 interestingly, the presence of pTT8 in the same cells improved the
 stability of pKMH052 at 70°C. Cloning of the crtB gene of *T.*
thermophilus HB27 encoding phytoene synthase into pKMH052, and
 introduction into *T. thermophilus* cells resulted in a 2.8-fold production
 of carotenoids, indicating the potential use of this plasmid for
 overexpression of genes from thermophiles and hyperthermophiles. .COPYRGT.
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ACCESSION NUMBER: 2004155468 EMBASE
TITLE: Fungal ammonia fermentation, a novel metabolic mechanism that couples the dissimilatory and assimilatory pathways of both nitrate and ethanol: Role of acetyl CoA synthetase in anaerobic ATP synthesis.
AUTHOR: Takasaki K.; Shoun H.; Yamaguchi M.; Takeo K.; Nakamura A.; Hoshino T.; Takaya N.
CORPORATE SOURCE: N. Takaya, Institute of Applied Biochemistry, University of Tsukuba, Tsukuba, Ibaraki 305-8572, Japan.
ntakaya@sakura.cc.tsukuba.ac.jp
SOURCE: Journal of Biological Chemistry, (26 Mar 2004) Vol. 279, No. 13, pp. 12414-12420.
Refs: 32
ISSN: 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 004 Microbiology
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 20040506
Last Updated on STN: 20040506
AB Fungal ammonia fermentation is a novel dissimilatory metabolic mechanism that supplies energy under anoxic conditions. The fungus *Fusarium oxysporum* reduces nitrate to ammonium and simultaneously oxidizes ethanol to acetate to generate ATP (Zhou, Z., Takaya, N., Nakamura, A., Yamaguchi, M., Takeo, K., and Shoun, H. (2002) J. Biol. Chemical 277, 1892-1896). We identified the *Aspergillus nidulans* genes involved in ammonia fermentation by analyzing fungal mutants. The results showed that assimilatory nitrate and nitrite reductases (the gene products of *niaD* and *niiA*) were essential for reducing nitrate and for anaerobic cell growth during ammonia fermentation. We also found that ethanol oxidation is coupled with nitrate reduction and catalyzed by alcohol dehydrogenase, coenzyme A (CoA)-acylating aldehyde dehydrogenase, and acetyl-CoA synthetase (Acs). This is similar to the mechanism suggested in *F. oxysporum* except *A. nidulans* uses Acs to produce ATP instead of the ADP-dependent acetate kinase of *F. oxysporum*. The production of Acs requires a functional *facA* gene that encodes Acs and that is involved in ethanol assimilation and other metabolic processes. We purified the gene product of *facA* (FacA) from the fungus to show that the fungus acetylates FacA on its lysine residue(s) specifically under conditions of ammonia fermentation to regulate its substrate affinity. Acetylated FacA had higher affinity for acetyl-CoA than for acetate, whereas non-acetylated FacA had more affinity for acetate. Thus, the acetylated variant of the FacA protein is responsible for ATP synthesis during fungal ammonia fermentation. These results showed that the fungus ferments ammonium via coupled dissimilatory and assimilatory mechanisms.

ACCESSION NUMBER: 2003462224 EMBASE
TITLE: Involvement of poly(ADP-ribose) synthetase in acoustic trauma of the cochlea.
AUTHOR: Tabuchi K.; Hoshino T.; Murashita H.; Oikawa K.; Uemaetomari I.; Nishimura B.; Tobita T.; Hara A.
CORPORATE SOURCE: K. Tabuchi, Department of Otolaryngology, Institute of Clinical Medicine, University of Tsukuba, 1-1-1 Tennodai, Tsukuba 305-8575, Japan. ktabuchi@md.tsukuba.ac.jp
SOURCE: Tohoku Journal of Experimental Medicine, (2003) Vol. 200, No. 4, pp. 195-202.
Refs: 24
ISSN: 0040-8727 CODEN: TJEMAO
COUNTRY: Japan
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 011 Otorhinolaryngology
029 Clinical Biochemistry

030 Pharmacology
037 Drug Literature Index

LANGUAGE: English
SUMMARY LANGUAGE: English
ENTRY DATE: Entered STN: 20031201
Last Updated on STN: 20031201

AB We investigated effects of poly(ADP-ribose) synthetase (PARS) inhibitors on acoustic trauma. Albino guinea pigs were intravenously given 3-aminobenzamide, nicotinamide or 3-aminobenzoic acid (an inactive analog of 3-aminobenzamide) just prior to exposure to a 2 kHz pure tone of 120 dB sound pressure level (SPL) for 10 minutes. The threshold of the compound action potential (CAP) and the amplitude of distortion-product otoacoustic emissions (DPOAEs) were measured before and 4 hours after the acoustic overexposure. Statistically significant decreases in the CAP threshold shifts and significant increases in the DPOAE amplitudes were observed 4 hours after the acoustic overexposure in the animals treated with 3-aminobenzamide or nicotinamide, whereas 3-aminobenzoic acid did not exert any protective effect. These results strongly suggest that excessive activation of PARS is involved in generation of the acoustic trauma. .COPYRG. 2003 Tohoku University Medical Press.

L29 ANSWER 4 OF 13 MEDLINE on STN

ACCESSION NUMBER: 2002097803 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11827254
TITLE: Differential and quantitative analyses of mRNA expression of glucosyltransferases from Streptococcus mutans MT8148.
AUTHOR: Fujiwara T; Hoshino T; Ooshima T; Hamada S
CORPORATE SOURCE: Department of Pedodontics, Osaka University Graduate School of Dentistry, Yamadaoka, Suita, Japan.. fujiwara@dent.osaka-u.ac.jp
SOURCE: Journal of dental research, (2002 Feb) 81 (2) 109-13.
Journal code: 0354343. ISSN: 0022-0345.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Dental Journals; Priority Journals
ENTRY MONTH: 200202
ENTRY DATE: Entered STN: 20020206
Last Updated on STN: 20020222
Entered Medline: 20020221

AB Streptococcus mutans produces three glucosyltransferases, coded by gtfB, gtfC, and gtfD, whose cooperative action is essential for sucrose-dependent cellular adhesion. This cellular adhesion plays an important role in the formation of dental plaque and the initiation of dental caries. Since they bear genetic similarities and are large in size, differentiation of their gene expression has been difficult, and little is known about the dynamic process of gtf expression. Using a real-time reverse-transcription/polymerase chain-reaction, we determined the expression of each gtf. Under various conditions, the relative levels of transcription were gtfB > gtfD > gtfC. Sucrose enhanced gtfD expression, whereas it reduced that of gtfB and gtfC, suggesting the presence of independent promoters. Quantitative analyses demonstrated coincidence between the ratio of expression of each gtf and the ratio previously identified as optimal for sucrose-dependent adhesion in vitro, suggesting that S. mutans produces GTF at an optimal ratio to adhere to the tooth surface.

L29 ANSWER 5 OF 13 MEDLINE on STN

ACCESSION NUMBER: 2001550735 MEDLINE
DOCUMENT NUMBER: PubMed ID: 11597030
TITLE: Contributions of three glycosyltransferases to sucrose-dependent adherence of Streptococcus mutans.
AUTHOR: Ooshima T; Matsumura M; Hoshino T; Kawabata S; Sobue S; Fujiwara T
CORPORATE SOURCE: Department of Pedodontics, Osaka University Graduate School of Dentistry, Suita, Japan.. ooshima@dent.osaka-u.ac.jp
SOURCE: Journal of dental research, (2001 Jul) 80 (7) 1672-7.
Journal code: 0354343. ISSN: 0022-0345.
PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Dental Journals; Priority Journals
ENTRY MONTH: 200110
ENTRY DATE: Entered STN: 20011015
Last Updated on STN: 20011029
Entered Medline: 20011025

AB Streptococcus mutans produces 3 types of glucosyltransferase (GTF), whose cooperative action is considered to be essential for its cellular adherence to the tooth surface. However, the precise mechanisms for synthesizing adhesive glucans and the specific roles of each GTF in cellular adherence to smooth surfaces have not been elucidated. In the present study, seven types of isogenic mutants of S. mutans MT8148 lacking GTFB, GTFC, and/or GTFD activities were constructed by inactivation of the genes encoding GTFB, GTFC, and/or GTFD. Furthermore, recombinant GTFB, GTFC, and GTFD were prepared from Escherichia coli cells harboring recombinant plasmids containing each of the gtf genes. Using these GTF-deficient mutants and rGTFs, we reconstituted sucrose-dependent adherence of S. mutans resting cells and examined the role of each GTF in vitro. The highest level of sucrose-dependent adherence was found at the ratio of 20 rGTFB:1 rGTFC:4 rGTFD in both the resting cells of GTF-deficient mutants and insoluble glucan synthesized by rGTFs. Moreover, when rGTFC and rGTFD were both present at concentrations of 1.5 mU and 6 mU, respectively, the insoluble glucan synthesized from sucrose by the rGTFs showed a high level of adhesiveness to smooth surfaces, even without rGTFB. These results suggest that the presence of all three GTFs at the optimum ratio is necessary for sucrose-dependent adherence of S. mutans, and that GTFC and GTFD may play significant roles in the synthesis of adhesive and insoluble glucan from sucrose.

L29 ANSWER 6 OF 13 MEDLINE on STN
ACCESSION NUMBER: 2000245547 MEDLINE
DOCUMENT NUMBER: PubMed ID: 10781558
TITLE: Contribution of cysteine desulfurase (NifS protein) to the biotin synthase reaction of Escherichia coli.
AUTHOR: Kiyasu T; Asakura A; Nagahashi Y; Hoshino T
CORPORATE SOURCE: Department of Applied Microbiology, Nippon Roche Research Center, 200 Kajiwara, Kamakura, Kanagawa 247-8530, Japan.. tatsuya.kiyasu@roche.com
SOURCE: Journal of bacteriology, (2000 May) 182 (10) 2879-85.
Journal code: 2985120R. ISSN: 0021-9193.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200006
ENTRY DATE: Entered STN: 20000622
Last Updated on STN: 20021008
Entered Medline: 20000612

AB The contribution of cysteine desulfurase, the NifS protein of Klebsiella pneumoniae and the IscS protein of Escherichia coli, to the biotin synthase reaction was investigated in in vitro and in vivo reaction systems with E. coli. When the nifS and nifU genes of K. pneumoniae were coexpressed in E. coli, NifS and NifU proteins in complex (NifU/S complex) and NifU monomer forms were observed. Both the NifU/S complex and the NifU monomer stimulated the biotin synthase reaction in the presence of L-cysteine in an in vitro reaction system. The NifU/S complex enhanced the production of biotin from dethiobiotin by the cells growing in an in vivo reaction system. Moreover, the IscS protein of E. coli stimulated the biotin synthase reaction in the presence of L-cysteine in the cell-free system. These results strongly suggest that cysteine desulfurase participates in the biotin synthase reaction, probably by supplying sulfur to the iron-sulfur cluster of biotin synthase.

L29 ANSWER 7 OF 13 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN
ACCESSION NUMBER: 2000:34013 SCISEARCH
THE GENUINE ARTICLE: 270RU
TITLE: A prokaryotic gene cluster involved in synthesis of lysine

through the amino adipate pathway: A key to the evolution of amino acid biosynthesis

AUTHOR: Nishida H; Nishiyama M (Reprint); Kobashi N; Kosuge T; Hoshino T; Yamane H
CORPORATE SOURCE: Univ Tokyo, Biotechnol Res Ctr, Tokyo 1138657, Japan (Reprint); Univ Tokyo, Inst Mol & Cell Biosci, Tokyo 1130032, Japan; Univ Tsukuba, Inst Appl Biochem, Tsukuba, Ibaraki 3058572, Japan
COUNTRY OF AUTHOR: Japan
SOURCE: GENOME RESEARCH, (DEC 1999) Vol. 9, No. 12, pp. 1175-1183. ISSN: 1088-9051.
PUBLISHER: COLD SPRING HARBOR LAB PRESS, 1 BUNG TOWN RD, PLAINVIEW, NY 11724 USA.
DOCUMENT TYPE: Article; Journal
LANGUAGE: English
REFERENCE COUNT: 33
ENTRY DATE: Entered STN: 2000
Last Updated on STN: 2000

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB In previous studies we determined the nucleotide sequence of the gene cluster containing lys20, hacA (lys4A), hacB (lys4B), orfE, orfF, rimK, argC, and argB of *Thermus thermophilus*, an extremely thermophilic bacterium. In this study, we characterized the role of each gene in the cluster by gene disruption and examined auxotrophy in the disruptants. All disruptants except for the orfE disruption showed a lysine auxotrophic phenotype. This was surprising because this cluster consists of genes coding for unrelated proteins based on their names, which had been tentatively designated by homology analysis. Although the newly found pathway contains alpha-amino adipic acid as a lysine biosynthetic intermediate, this pathway is not the same as the eukaryotic one. When each of the gene products was phylogenetically analyzed, we found that genes evolutionarily related to the lysine biosynthetic genes in *T. thermophilus* were all present in a hyperthermophilic and anaerobic archaeon, *Pyrococcus horikoshii*, and formed a gene cluster in a manner similar to that in *T. thermophilus*. Furthermore, this gene cluster was analogous in part to the present leucine and arginine biosynthesis pathways. This lysine biosynthesis cluster is assumed to be one of the origins of lysine biosynthesis and could therefore become a key to the evolution of amino acid biosynthesis.

L29 ANSWER 8 OF 13 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1999:533591 SCISEARCH

THE GENUINE ARTICLE: 217KZ

TITLE: Cloning and sequencing of clustered genes involved in fatty acid biosynthesis from the docosahexaenoic acid-producing bacterium, *Vibrio marinus* strain MP-1

AUTHOR: Morita N; Ueno A; Tamaka M; Ohgiya S; Hoshino T; Kawasaki K; Yumoto I; Ishizaki K; Okuyama H (Reprint)

CORPORATE SOURCE: Hokkaido Natl Ind Res Inst, AIST MITI, Biosci & Chem Div, Toyohira Ku, Sapporo, Hokkaido 0628517, Japan (Reprint); Hokkaido Univ, Grad Sch Environm Earth Sci, Lab Environm Mol Biol, Kita Ku, Sapporo, Hokkaido 0600810, Japan

COUNTRY OF AUTHOR: Japan

SOURCE: BIOTECHNOLOGY LETTERS, (JUL 1999) Vol. 21, No. 7, pp. 641-646.

ISSN: 0141-5492.

PUBLISHER: KLUWER ACADEMIC PUBL, VAN GODEWIJCKSTRAAT 30, 3311 GZ DORDRECHT, NETHERLANDS.

DOCUMENT TYPE: Article; Journal

LANGUAGE: English

REFERENCE COUNT: 19

ENTRY DATE: Entered STN: 1999

Last Updated on STN: 1999

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB A cluster of genes involved in fatty acid biosynthesis (fab) was isolated from docosahexaenoic acid (DHA)-producing *Vibrio marinus* strain MP-1. This fab gene cluster included five genes highly homologous to the *Escherichia coli* counterparts, and their order in the cluster was the same

with that of the E. coli fab gene cluster except that the latter included the additional fabH gene. These fab genes should be involved in early steps of DHA biosynthesis in V. marinus strain MP-1.

L29 ANSWER 9 OF 13 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1996:150043 SCISEARCH

THE GENUINE ARTICLE: TW477

TITLE: Mapping of 61 genes on the refined physical map of the chromosome of Thermus thermophilus HB27 and comparison of genome organization with that of T-thermophilus HB8

AUTHOR: Tabata K (Reprint); Hoshino T

CORPORATE SOURCE: UNIV TSUKUBA, INST APPL BIOCHEM, TSUKUBA, IBARAKI 305, JAPAN

COUNTRY OF AUTHOR: JAPAN

SOURCE: MICROBIOLOGY-UK, (FEB 1996) Vol. 142, Part 2, pp. 401-410. ISSN: 1350-0872.

PUBLISHER: SOC GENERAL MICROBIOLOGY, HARVEST HOUSE 62 LONDON ROAD, READING, BERKS, ENGLAND RG1 5AS.

DOCUMENT TYPE: Article; Journal

FILE SEGMENT: LIFE

LANGUAGE: English

REFERENCE COUNT: 54

ENTRY DATE: Entered STN: 1996

Last Updated on STN: 1996

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB We have constructed refined physical maps of the chromosome (1.82 Mb) and the large plasmid pTT27 (250 kb) of Thermus thermophilus HB27. A total of 49 cleavage sites with five restriction enzymes, EcoRI, SspI, MunI, EcoRV and ClaI, were determined on the maps. The location of 61 genes was determined by using as probes 64 genes cloned from T. thermophilus or other Thermus strains. Comparison of the genomic organization of the chromosomes of T. thermophilus HB27 and HB8 revealed that they were basically identical, but some genes were located in different regions. Among 32 genes whose locations were determined on both the HB27 and the HB8 chromosomes, the copy number of rpsL-rpsG-fus-tufA, the locations of glyS, pol, and one copy of nusC-rplK-rpA were different. The 151000 sequence was located only in one region on the HB27 chromosome. In contrast, 151000 sequences were scattered over four regions on the chromosome of HB8. As each region in which glyS, pol, or one copy of nusG-rplK-rplA are present also contained 151000 in HB8, it is suggested that 151000 may play an important role in genomic rearrangements in Thermus strains.

L29 ANSWER 10 OF 13 MEDLINE on STN DUPLICATE 3

ACCESSION NUMBER: 94354585 MEDLINE

DOCUMENT NUMBER: PubMed ID: 8074461

TITLE: Relationship between cellular glutathione level and susceptibility to LAK killing in human pharyngeal carcinoma cell line.

AUTHOR: Noda I; Fujieda S; Saito H; Tanaka N; Sugimoto C; Hoshino T; Yagita M

CORPORATE SOURCE: Department of Oto-Rhino-Laryngology, Fukui Medical School, Japan.

SOURCE: Anticancer research, (1994 May-Jun) 14. (3A) 1117-20. Journal code: 8102988. ISSN: 0250-7005.

PUB. COUNTRY: Greece

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199409

ENTRY DATE: Entered STN: 19941006

Last Updated on STN: 19941006

Entered Medline: 19940926

AB We examined the relationship between cellular glutathione (GSH) level and susceptibility to lymphokine-activated killer (LAK) cell-mediated cytotoxicity in KB human pharyngeal carcinoma cells. Treatment of KB cells with D,L-buthionine-S,R-sulfoximine (BSO), a gamma-glutamyl cysteine synthetase blocker, resulted in decreased total intracellular GSH

levels associated with increased susceptibility to LAK killing. In contrast, treatment with oxothiazolidine-4-carboxylate (OTZ, a precursor of cysteine), which is known to increase cellular GSH level, decreased the susceptibility of KB cells to LAK killing. Both agents had no effects on binding frequency of KB cells to LAK cells. These results suggest that intracellular GSH in tumor cells play a protective role against LAK mediated cytolysis, specially in the post-binding killing phase.

L29 ANSWER 11 OF 13 EMBASE COPYRIGHT (c) 2005 Elsevier B.V. All rights reserved on STN DUPLICATE 4

ACCESSION NUMBER: 84191025 EMBASE
DOCUMENT NUMBER: 1984191025
TITLE: Experimental combination chemotherapy with thymidylate synthetase and ribonucleotide reductase inhibitors.
AUTHOR: Kobayashi S.; Hoshino T.; Santi D.V.
CORPORATE SOURCE: Brain Tumor Research Center, University of California, San Francisco, CA 94143, United States
SOURCE: Pharmaceutical Research, (1984) Vol. NO. 4, pp. 181-183.
CODEN: PHREEB
COUNTRY: Germany
DOCUMENT TYPE: Journal
FILE SEGMENT: 037 Drug Literature Index
030 Pharmacology
016 Cancer
LANGUAGE: English
ENTRY DATE: Entered STN: 911210
Last Updated on STN: 911210

L29 ANSWER 12 OF 13 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1983:563593 SCISEARCH
THE GENUINE ARTICLE: RN325
TITLE: EXPERIMENTAL CHEMOTHERAPY WITH THYMIDYLATE SYNTHETASE AND RIBONUCLEOTIDE REDUCTASE INHIBITORS
AUTHOR: HOSHINO T (Reprint); KOBAYASHI S; SANO Y
CORPORATE SOURCE: UNIV CALIF SAN FRANCISCO, SAN FRANCISCO, CA 94143
COUNTRY OF AUTHOR: USA
SOURCE: CELL AND TISSUE KINETICS, (1983) Vol. 16, No. 6, pp. 615-615.
ISSN: 0008-8730.
PUBLISHER: BLACKWELL PUBLISHING LTD, 9600 GARSINGTON RD, OXFORD OX4 2DG, OXON, ENGLAND.
DOCUMENT TYPE: Conference; Journal
LANGUAGE: English
REFERENCE COUNT: 0
ENTRY DATE: Entered STN: 1994
Last Updated on STN: 1994

L29 ANSWER 13 OF 13 SCISEARCH COPYRIGHT (c) 2005 The Thomson Corporation on STN

ACCESSION NUMBER: 1983:603800 SCISEARCH
THE GENUINE ARTICLE: RM382
TITLE: CHEMOTHERAPY WITH THYMIDYLATE SYNTHETASE AND RIBONUCLEOTIDE REDUCTASE INHIBITORS
AUTHOR: HOSHINO T (Reprint); KOBAYASHI S; SANO Y
CORPORATE SOURCE: UNIV CALIF SAN FRANCISCO, SAN FRANCISCO, CA 94143
COUNTRY OF AUTHOR: USA
SOURCE: PROCEEDINGS OF THE AMERICAN ASSOCIATION FOR CANCER RESEARCH, (1983) Vol. 24, No. MAR, pp. 298-298.
ISSN: 0197-016X.
PUBLISHER: AMER ASSOC CANCER RESEARCH, 615 CHESTNUT ST, 17TH FLOOR, PHILADELPHIA, PA 19106-4404 USA.
DOCUMENT TYPE: Conference; Journal
LANGUAGE: English
REFERENCE COUNT: 0
ENTRY DATE: Entered STN: 1994
Last Updated on STN: 1994

=> d his

(FILE 'HOME' ENTERED AT 14:02:11 ON 04 NOV 2005)

FILE 'MEDLINE, EMBASE, BIOSIS, BIOTECHDS, SCISEARCH, HCAPLUS, NTIS, LIFESCI' ENTERED AT 14:02:36 ON 04 NOV 2005

L1 4 S ASTAXANTHIN (W)SYNTHASE?
L2 0 S ASTAXANTHIN (W)SYNTHETASE?
L3 6416 S ASTAXANTHIN
L4 24046 S "AST"
L5 194127 S SYNTHETASE?
L6 0 S L3 (W)L5
L7 39 S L3 AND L5
L8 24 DUP REM L7 (15 DUPLICATES REMOVED)
L9 41050 S REACTION (A) CENTER
L10 0 S L8 AND L9
L11 153 S L5 AND L9
L12 371127 S REDUCTASE?
L13 21 S L11 AND L12
L14 19 DUP REM L13 (2 DUPLICATES REMOVED)
L15 7593 S P450 REDUCTASE?
L16 0 S L9 AND L15
L17 0 S L4 AND L9
L18 59612 S "BETA-CAROTENE"
L19 64 S L4 AND L18
L20 0 S L9 AND L19
L21 0 S L1 AND L9
L22 3 DUP REM L1 (1 DUPLICATE REMOVED)
L23 93 DUP REM L11 (60 DUPLICATES REMOVED)
E HOSHINO T/AU
L24 4600 S E3
E KAZUYUKI O/AU
L25 6 S E3
E YUTAKA S/AU
L26 12 S E3
L27 4617 S L24 OR L25 OR L26
L28 19 S L5 AND L27
L29 13 DUP REM L28 (6 DUPLICATES REMOVED)

	L #	Hits	Search Text
1	L1	0	astaxanthin adj synthetase?
2	L2	0	AST adj synthetase\$2
3	L3	2	astaxanthin adj synthetase\$3
4	L4	6391	beta adj carotene
5	L5	1541 8	synthetase\$2
6	L6	130	l4 same l5
7	L7	7843 08	clon\$3 or express\$3 or recombinant
8	L8	29	l6 same l7
9	L9	6955	HOSHINO KAZAYUKI yukata
10	L10	1	l6 and l9

	Issue Date	Page s	Document ID	Title
1	20040101	77	US 2004000343 0 A1	4-ketocarotenoids in flower petals
2	20020402	32	US 6365386 B1	Astaxanthin synthase

	Issue Date	Page s	Document ID	Title
1	20051027	59	US 2005023976 9 A1	Therapeutic compounds for treating dyslipidemic conditions
2	20050728	39	US 2005016503 9 A1	Fluorinated 4-azasteroid derivatives as androgen receptor modulators
3	20050616	73	US 2005013100 5 A1	4-azasteroid derivatives as androgen receptor modulators
4	20050602	50	US 2005011934 3 A1	Dihydroxy open-acid salt of simvastatin
5	20050526	37	US 2005011341 9 A1	Therapeutic compounds for treating dyslipidemic conditions
6	20050519	61	US 2005010741 6 A1	17-hydroxy 4-aza androstan -3-ones as androgen receptor modulators
7	20050120	38	US 2005001480 7 A1	Therapeutic compounds for treating dyslipidemic conditions
8	20050120	51	US 2005001421 9 A1	Genes encoding carotenoid compounds
9	20041230	47	US 2004026843 9 A1	Genes encoding carotenoid compounds
10	20041230	29	US 2004026684 9 A1	Therapeutic compounds for treating dyslipidemic conditions
11	20041216	49	US 2004025366 3 A1	Genes encoding carotenoid compounds

12	20041125	33	US 2004023580 8 A1	Androstanes as androgen receptor modulators
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	Issue Date	Page s	Document ID	Title
13	20041118	33	US 2004022984 4 A1	Method of treating atherosclerosis, dyslipidemias and related conditions
14	20041104	37	US 2004022015 9 A1	Androstane 17-beta-carboxamides as androgen receptor modulators
15	20041007	36	US 2004019871 7 A1	Androgen receptor modulators and methods of use thereof
16	20040916	20	US 2004018039 2 A1	Screening and selection methods for statin drug combinations
17	20040617	11	US 2004011651 0 A1	Antihypertensive agent and cholesterol absorption inhibitor combination therapy
18	20040122	146	US 2004001404 0 A1	Cardiotoxin molecular toxicology modeling
19	20031113	23	US 2003021115 1 A1	Dihydroxy open-acid and salts of HMG-Co-A reductase inhibitors
20	20030918	54	US 2003017650 1 A1	Dihydroxy open-acid salt of simvastatin
21	20030807	32	US 2003014831 9 A1	Genes encoding carotenoid compounds
22	20030703	33	US 2003012535 7 A1	Therapeutic compounds for treating dyslipidemic conditions
23	20030619	44	US 2003011563 9 A1	Expressed sequences of arabidopsis thaliana
24	20030508	16	US 2003008692 3 A1	Method for the prevention and/or treatment of atherosclerosis

	Issue Date	Page s	Document ID	Title
25	20030403	103	US 2003006500 4 A1	Androgen receptor modulators and methods for use thereof
26	20050816	47	US 6929928 B2	Genes encoding carotenoid compounds
27	20050621	31	US 6908934 B2	Therapeutic compounds for treating dyslipidemic conditions
28	20031111	71	US 6645974 B2	Androgen receptor modulators and methods for use thereof
29	20030527	23	US 6569461 B1	Dihydroxy open-acid and salts of HMG-CoA reductase inhibitors

	Issue Date	Page s	Document ID	Title
1	20020402	32	US 6365386 B1	Astaxanthin synthase